ENHANCING THE ORGANOLEPTIC QUALITIES OF PASTEURIZED AND IN-CONTAINER STERILIZED MILK.

By

A.M.I.Wijerathna

(03/AS/083)

The thesis is submitted in partial fulfillment of the requirement for the

Special Degree of Bachelor of Sciences

In

Food Science & Technology

Department of Food Science & Technology

Faculty of Applied Sciences

Sabaragamuwa University of Sri Lanka

Belihuloya

March 2009

DECLARATION

The work describe in this thesis was carried out by me at the Department of Food Science & Technology, Faculty of Applied Sciences, Sabaragamuwa University of Sri Lanka, under the supervision of Mr. A.I.C.J. Liyanage and Mr. A.R.V. Abesinghe. The report on this has not been submitted to another university for another degree.

A.M.I.Wijerathna (03/AS/083)

27.04.2009

Date

Certified by:

External Supervisor,

Mr. A.R.V. Abesinghe
Assit. Manager - Production
Milk Industry of Lanka Company (MILCO) Ltd.
Rajawella
Digana.

Signature

27.04.2009.

Date

Internal Supervisors,

Mr. A.I..C.J. Liyanage
Lecturer - Department of Food Science & Technology
Faculty of Applied Sciencees
Sabaragamuwa University of Sri Lanka
Belihuloya

Signature

29-04-2009

Date

The Head of the Department,

Mr. M.C.N. Jayasooriya
The Head, Department of Food Science & Technology
Faculty of Applied Sciences
Sabaragamuwa University of Sri Lanka
Belihuloya

Signature

29/04/09

Date

Head/Dept. of Food Science & Technology
Faculty of Applied Sciences
Sabaragamuwa University of Sri Lanka
Belihuloya

ACKNOWLEDGEMENT

I am deeply indebted to my external supervisor Mr. A.R.V. Abesinghe and Mr. A.L.C.J. Liyanage for invaluable guidance, encouragement, supervision and co-ordination given during the period of my research work and preparation of thesis, because otherwise this work would not be a success.

I would like to express my heartiest thanks to Mr. M.C.N. Jayasooriya , actg.head/ Dept. of Food science & Technology and Mr. R.M.J.B. Ratnayake , factory manager of Digana Milk Plant for providing me to this opportunity to carry out this study at MILCO (pvt) ltd.

Finally, I would like to express my sincere thanks to library staff of Faculty of Applied Sciences and staff members in MILCO, who gave me a great support and helped me in many ways during the research.

ABSTRACT

Cocoa powder is used to produce in-container sterilized and pasteurized milk. Sedimentation of cocoa powder can be observed in in-container sterilized milk. The problem leads to reduce the quality of pasteurized and in-container sterilized milk. Hence this study was aimed to improve the organoleptic quality of stabilizers to control this problem. Simultaneously other organoleptic qualities were also aimed to enhance by using chocolate liquid. The study was carried out at MILCO (Pvt) Ltd, Digana.

The amount of sediment for each product was quantified using oven drying method. In order to choose a stabilizer, trials were carried out using Carrageenan, Sodium citrate and a combination of Carrageenan and Sodium citrate to reduce the amount of sediment in in – container sterilized milk. Finally a suitable amount of the stabilizer- "Carrageenan" was estimated. It was 0.01%.

Three in-container sterilized milk samples were prepared by changing the level of chocolate liquid as 90%, 60%, 30% and cocoa powder as 10%, 40%, 70%, for each sample respectively and level of Carrageenan was 0.01% for each, while keeping the other ingredients constant. Three pasteurized milk samples were prepared by changing the level of chocolate liquid as 20%, 40%, 60% and cocoa powder as 80%, 60%, 40% while keeping the other ingredients constant. Sensory evaluation was carried out for three recipes formulated for the above two products, with a panel consisting thirty non-trained panelists to select best sample.

Analyzed results of the final product in-container sterilized milk were 2.0%of fat, 0.15 acidity, and 17.6% of total solids Analyzed results of the final product of pasteurized milk were 2.1%of fat, 0.15 acidity, 17.8% total solids. The observed total colony count for pasteurized milk was 30000 per /ml and methythlene blue dye reduction was observed after 4 hours.

The data obtained by the sensory evaluation were statistically analyzed using Friedman test at 5%significance level. According to the results the best sample of in-container sterilized milk was selected as the one containing chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant. According to the amount of sedimentation above sample was the best one. The sensory data obtained for pasteurized milk samples were statistically analyzed using the same procedure and the sample containing 20% chocolate liquid and 80% cocoa powder was selected as the best one.According to the product specification Carrageenan 0.01% was selected as suitable amount to effectively reduce the sediment in in-container sterilized milk. The reduction in sediment achieved was approximately 65%.

CONTENTS

CHAPTER 03: MATERIALS AND METHODOLOGY

CHAPTER 04: RESULTS AND DISCUSSION

LIST OF ABREVIATIONS

cal	:Calorie
°C	:Centigrade
FAO	:Food and Agriculture Organization
g	:Grams
HTST	:High Temperature Short Time
hrs	:Hours
M	: Mole
mcg	:Micrograms
Min	:Minutes
mg	:Milligrams
ml	:Milliliters
Nos	:Numbers
ppm	:Parts per million
rpm	:Round per minute
s	:Seconds
SLS	:Sri Lanka Standard
TCC	:Total Colony Count
TPC	:Total Plate Count
UHT	:Ultra Heat Treatment
w/w	:Weight to weight
WHO	:World Health Organization

LIST OF FIGURES

LIST OF TABLES

Tables **Pages**

CHAPTER 01

INTRODUCTION

1.1. Background:

Milk Industry of Lanka Company Ltd. (MILCO) is a leading company in Sri Lanka, engaged in the process of manufacturing range of milk based products including milk powder, butter, condensed milk, sterilized milk, pasteurized milk, cheese, yoghurt, ghee etc. and marketed under the brand name "Highland". It is proud to be a Sri Lankan company to manufacture their dairy products with a Sri Lankan identity. Chocolate in - container sterilized and pasteurized milk has ,since its introduction by MILCO (Pvt)ltd , been very popular especially among children and adults and is often used to introduce milk products, where fresh milk has been relatively unknown.

Cocoa powder is used to produce in-container sterilized and pasteurized milk. Sedimentation of cocoa powder can be observed in-container sterilized milk. The problem leads to reduce the quality of pasteurized and in-container sterilized milk. It has been realized that lack of satisfactory of consumers. Depending on the type of cocoa powder used, viscosity can vary substantially. Only part of the cocoa powder will dissolve in the milk, the majority of the particles settling out as sediment over a period of time. So, this reason cause to reduce organoleptic qualities of pasteurized and in-container sterilized milk.

According to this problem, consumer acceptability of quality demand for "Highland" in container sterilized milk and pasteurized milk make a relatively lower in present market dynamics. To be ahead in the market it is indispensable to enhance overall sensory attributes up to the level in which consumer utmost satisfaction prevails. Hence this study was aimed to improve the organoleptic quality of stabilizers to control this problem. Simultaneously other organoleptic qualities were also aimed to enhance by using chocolate liquid.

There are so many stabilizers. Stabilizers are added in order to avoid sediment formation But ,it should be selected according to the type of product. "Carrageenan" and "Sodium Citrate" are example for stabilizers which can be used to avoid sedimentation.. The amount of stabilizer to be added should be the minimum required to avoid sedimentation. Carrageenan is a wholly natural ingredient obtained from certain species of the red seaweed, class *Rhodophyceae*

It consists of varying amounts of the ammonium, Calcium,Magnisium,Potasium or Sodium salts of sulphate esters of galactose and 3,6-anhydrogalactose copolmers are designated kappa, lambda,and iota .These three types differ in structure and their gelling ability.

Sodium salts of citric acid, a compound found in every living organism, as it is part of the key metabolic pathways in all body cells. Large concentrations are found in citrus fruits, kiwi, strawberries and many other fruits. Commercially prepared by fermentation of molasses with the mould *Aspergillus niger*.

Chcolate liquid named "L.i Co" used to enhance other organoleptic qualities in container sterilized milk and pasturezied milk

1.2. Overall Objective:

- Increasing the consumer preference of pasteurized and in-container sterilized milk products of MILCO (Pvt) Ltd by enhancing their organoleptic qualities.

1.3. Specific Objectives:

- To study the organoleptic qualities of pasteurized and in-container sterilized milk.
- To study the limitations for organoleptic qualities
- To increase the consumer preference by providing solutions for the above limitations.

CHAPTER 02

LITERATURE REVIEW

2.1. Milk

Milk is an opaque white liquid produced by the mammary glands of female mammals. It provides the primary source of nutrition for newborn mammals before they are able to digest other types of food. Cow milk was first used as human food in the Middle East. Milk was first delivered in bottles in1878 .Technology allows processors to disassemble milk into its various component parts (Figure2.1.)and these can be combined in different ways to make new products or used as specific ingredients in other foods. Approximately 80% of total world milk delivered to milk factories in 1993, was processed into products. This shows the importance of milk composition to the processor. Milk composition is also of extreme importance to the consumer from a nutrition point of view. In this health conscious age, changing the ratio of protein to fat or indeed changing the fatty acid composition of the milk is now perceived as being desirable. It should be remembered that changes in the diet of cows may influence the yield of constituents as well as their concentrations in milk.

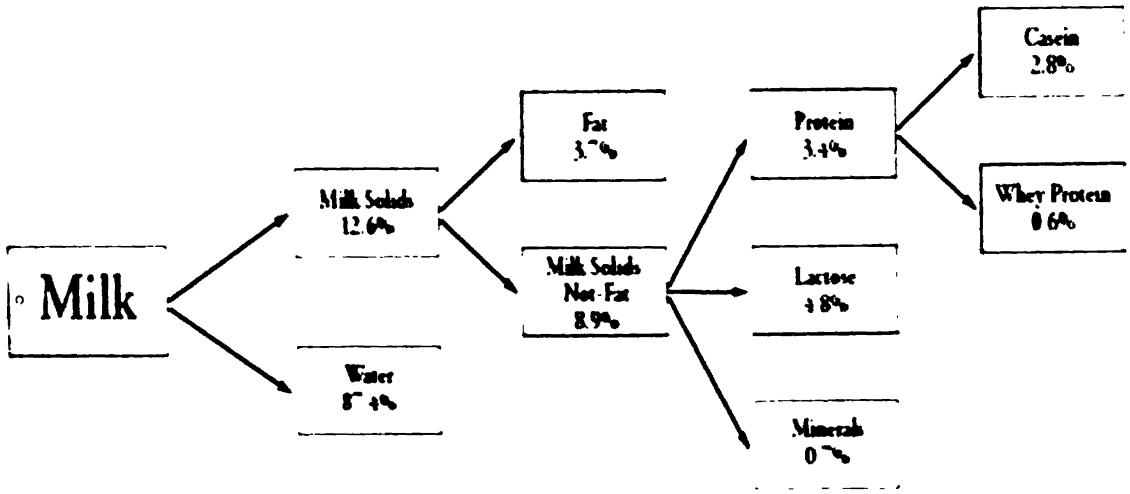


Figure 2.1. Constituents in milk (Chandan 1997)

2.2. Sterilized Milk

Strictly speaking, sterilized milk must be sterile that is it should contain neither bacteria nor bacterial spores, however, sometimes less stringent definitions are used that can be of practical value in many circumstances. Although, the theoretically correct definition of sterilized milk is one that must have ,at ambient temperatures, an unlimited keeping quality from the bacteriological point of view. This later definition does not always mean that sterilized milk must be completely free of bacterial spores. (FAO 1975)

2.2.1. Systems of Milk Sterilization

For the sterilization of milk several different methods are now in use,

1. In bottle sterilization

The milk is bottled and sterilized at temperatures between 105-120 °C.The sterilization can be done either in an autoclave (batch) or in a tower sterilizer (continuous).

2. Ultra High Temperature Short Time.

The milk is sterilized in a continuous-flow at very high temperature(130-150 °C) for very short time(1-20s).This method require aseptically packing in sterilized containers

3. Two stage process sterilization.

The milk is first sterilized according to the UHT process.Then bottled and finally submitted to further heat treatment to destroy any spore which may have entered during bottling.

2.2.2. Bacteriological Aspects of Milk Sterilization

When the raw milk is heat treated the spores are not immediately destroyed, their destruction is a process that need time, whether they are of different species or all of one specie. It has generally been accepted that spores, which heated at constant temperature are destroyed in such a manner that the relation between the logarithm of surviving

number and the time is presented by a straight line. Hence it called thermal destruction curve.

In dealing with sterilization process the terms "decimal reduction" and the "decimal reduction time" are often used. The first indicates reduction of the number of spores to one tenth of the original concentration the second, the time necessary for the performance of such a reduction. When spore destruction proceeds according to a straight curve the decimal reduction time is the same at all concentrations. From the course of thermal destruction curve it follows that the initial spore concentration has an important effect upon the result of heat treatment. (FAO 1975)

2.2.3. Chemical Aspects of Milk Sterilization

The higher temperature of the heat treatment, the greater the sterilization effect and the more marked the change in the colour and taste of the milk.When milk is heated off-flavours occur at first a cooked flavour, caused by the production of volatile sulfur compounds. When the intensity of the heat treatment is increased, a sterilization taste mainly caused by the reaction between the sugar the protein constituents (Millard reaction). This process influences not only the taste of the milk but also its colour, which become brownish. One important phenomenon is that with increasing temperature the spore destruction rate increases more than influences on the taste and colour of milk.With every 10°C increase the rate of the browning reaction multiplies approximately 2.5 fold, while the rate of spore-destruction is increased about tenfold. (Table 2.1.)

Table 2.1. The different temperatures has on the browning rate as compaired to the spore destruction rate.(Source:FAO 1975)

Temperature of heating (°C)	Relative spore destruction rate	Relative rate of browning reaction
100	1	1
110	10	2.5
120	100	6.2
130	1000	15.6

140	10000	39.0
150	100000	97.5

It becomes more apparent when studying the number of temperature time combinations in (Table2.1.) with the number same sterilizing effect and calculating the relative degree of browning caused by these heat treatments. All the time-temperature combination produce the same sterilizing effect.

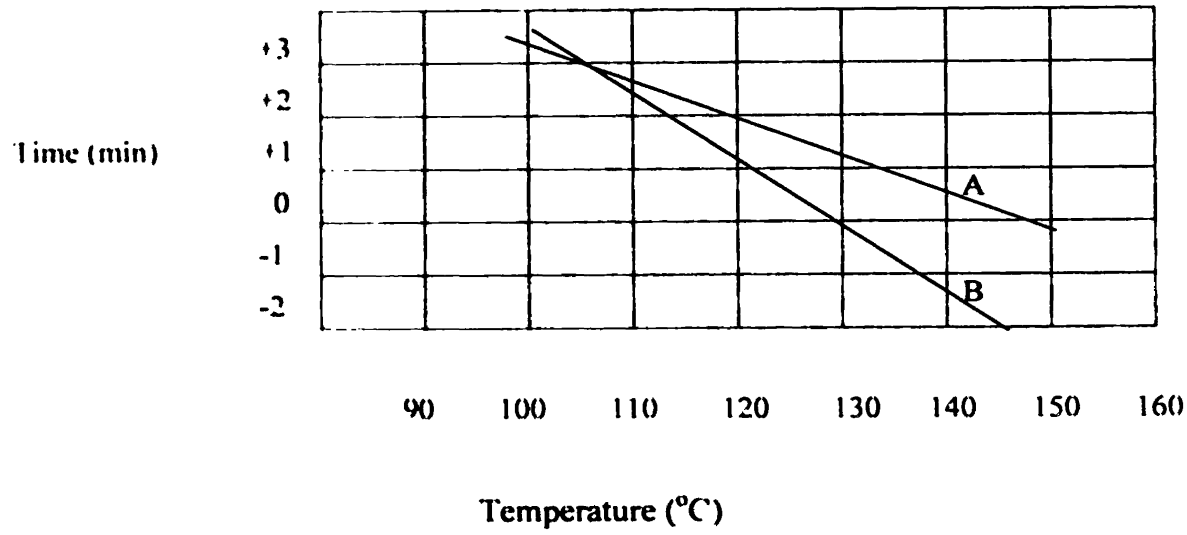


Figure 2.2. The time temperature curves for browning (A) and spore destruction (B) of milk.(Source:FAO 1975)

Figure 14 shows that at 110 °C the milk is sterile before it became brownish. This means that a sterile product is obtained that should be of normal colour. In practice, it is necessary to pay attention to the time of both heating and cooling. For proper sterilization there must be a certain safety margin in the duration of the heat treatment and this increases the possibilities of the heat induced faults described above.(FAO 1975)

2.3. Pasteurized Milk

International Dairy Federation defined pasteurized milk as,

A process applied to a product with the object the of minimizing possible health hazards arising from pathogenic microorganisms associated with milk by heat treatment, which is consistent with minimal chemical, physical and organoleptic changers in the product.(SDF 1983)

2.3.1. Pasteurization Methods

There are basically two types of methods for pasteurizing milk. They are Batch pasteurization and HTST (high temperature-short time) pasteurization. In batch pasteurizer the milk is heated to slightly above 63°C and held for at least 30 minutes. Batch pasteurizers have relatively small amount of heat exchange surface per kilo of product contained and therefore cool the milk slowly. Consequently, surface-type or plate cooler are commonly used to cool the milk to bottling temperature 4°C.HTST pasteurization is a continuous process in plate heat exchanger in which milk is rapidly heated to a temperature of not less than 72 °C and held for not less than 15 seconds and rapidly cooled down to a certain temperature depending on the size of regenerative and cooling sections.

2.4. Chocolate Milk

Chocolate has largely replaced cocoa powder for beverages. These drinks are easier to prepare and most of these chocolates can be used to make cold drinks. Chocolate milk has, since its introduction, been very popular especially among children and is often used to introduce milk products, where fresh milk has been relatively unknown. Many individuals that do not like plain milk will drink chocolate milk. It would appear that the addition of the syrups with its additional nutrients would increase the nutritive value of the milk.

In some countries the name chocolate milk can only be used when the fat percent is at least equal to that of minimum legal requirement for fluid milk. If the fat percent is lower

the product is called chocolate drinks. The compositions of chocolate milk vary very much depending upon the market situation and the availability of ingredients for the processing. In some countries, the legislation demands a minimum content of various. (Atherton and Newlander 1987)

Table2.2 Composition of sterilized milk and pasteurized milk (Source: Milk industry)

components	Pasteurized milk per 100g	Sterilized milk Per 100 g
Fat	2.00g	2.0g
Protein	3.40g	3.40g
carbohydrates	9.20g	11.60g
Vitamin A	52.20mcg	52.20mcg
Vitamin C	1.00mcg	-
Iron	0.20mg	0.20mg
Calcium	120mg	120mg
Phosphorous	90mg	30mg
Thiamin	50mg	50g
Riboflavin	190mcg	190mcg
Energy	73.106cal	76.99cal

2.5. Organoleptic Qualities

Fresh milk that has been produced under ideal conditions will have no pronounced flavor, but will have a slightly sweet and pleasant taste. This is primarily due to the relationship of the lactose and chloride contents. If this relationship is disturbed so that the chloride becomes relatively greater, as in late lactation or in masttitic conditions, the flavour is adversely affected. The flavor can be changed during the period of processing time of the product. The white colour of milk results from the dispersion of reflected light by the fat globules and the colloidal particles of casein and calcium phosphate. The yellow color is due to the pigment carotene which is fat soluble. Riboflavin is another pigment in milk.

When the milk is heated off flavors occur and the different types of heat treatments influences not only the taste of the milk but also its colour, which become brownish. Especially in-in container sterilized milk sediment of cocoa powder can be observed. Depending on the type of cocoa powder used, viscosity can vary substantially. Only part of the cocoa powder will dissolve in the milk, the majority of the particles settling out as sediment over a period of time. This visualizing sediment cause to reduce the quality in - container sterilized milk. (Atherton and Newlander 1987)

2.6. Basic Raw Materials

2.6.1. Milk

Milk is a dispersion of milk fat globules and casein micelles in a continuous phase of water, lactose, whey proteins, and minerals. Cow's milk is used predominantly in industrial chocolate milk production.

2.6.2. Cocoa Powder

Cocoa powder is the product obtained by mechanical transformation in to power of cocoa press cake which is the product obtained by mechanical transformation in to. The liquid character of chocolate milk brings out the full cocoa flavor components almost instantly. This contributes to why chocolate milk is so popular around the world. However, producing chocolate milk is a challenging task because only a part of the cocoa powder dissolves in the milk. The majority of the particles settle out as sediment over a period of time. Moreover, the processing method determines parameters such as alkalinity, which has great influence on the functional performance of cocoa powder Heat treatment influences the network contributing to viscosity and stability, because upon heating, cocoa solids bind to water, proteins and stabilizers. Depending on the type of cocoa powder used, viscosity can vary substantially. Cocoa powder color, flavor and pH are all variables that will affect the end product.

There are two types of cocoa powder. They are natural and alkalized cocoa powder. Appearance of them can be observed as homogenous powder, relatively free flowing, free of lumps and free of foreign materials no additive.

Table 2.3. Colours and components in natural and alkalized cocoa powder.(FAO 1975)

	Natural cocoa powder	Alkalized cocoa powder
Colour	Light brown	Dark brown
Moisture	6% maximum	5% maximum
Fat content	10-12%	10-12%
pH	5.0-5.6	6.8-7.2
Ash	9.0%	9.0%

Cocoa manufactured from substanded beans may be contaminated with insects fragments, mycotoxins and yeast. The larger manufactures who use cocoa as an ingredient in their products are becoming increasingly aware of its potential microbiological activity.

Table 2.4. Microbiological test for natural and alkalized cocoa powder (FAO 1975)

Microorganisms	Natural cocoa powder	Alkalized cocoa powder
Coliform	negative	negative
TCC	<1000 per g	<1000 per g
Yeast	-	Not more than 10 Nos per g
Moulds	-	Not more than 10 Nos per g

2.6.3. Sugar

In non-scientific use, the term sugar refers to sucrose a white crystalline solid disaccharide. In this informal sense, the word "sugar" principally refers to crystalline sugars.Humans most commonly use sucrose as their sugar of choice for altering the flavor and properties of beverages and food. Commercially produced table sugar comes either from sugar cane or from sugar beet.

2.7. Stabilizers

Stabilizer is a chemical which tends to inhibit the reaction between two or more other chemicals. It can be thought of as the antonym to a catalyst. It can be also a chemical that inhibits separation of suspensions, emulsions, and foams Stabilizers are added in order to avoid sediment formation. The milk's natural stabilizers (casein, albumin, and globulin) should be kept intact by avoiding too high pasteurization temperatures. The amount of stabilizer to be added should be the minimum required to avoid sedimentation. The viscosity in the chocolate milk will be too high if too much stabilizer is added. Common stabilizers are Carrageenan, Sodium Algenate, Agar and Gelatine.(FAO 1975)

2.7.1. Carrageenan

Figure 2.3. Red seaweed: *Chondrus crispus*

Carrageenan (E407)is a wholly natural ingredient obtained from certain species of the red seaweed, class *Rhodophyceae*. Popular sources for carrageenan are the *Chondrus Crispus*, *Eucheuma Cottonii* and *Eucheuma Spinosum* species. The *Chondrus Crispus* specie grows mainly in cold-water territories such as the northern coasts of the Atlantic while the *Eucheuma* species are abundantly found in temperate climates like the Philippines.

The seaweeds are dried mechnicaly or are left on the shore to be alternatively bleached and soaked, and then dried. They are then washed to remove salts and debries befor extraction process using alkaline hot water. The resulting dried mucilge is translucent and swells in cold water, partially dissolving to produce a jelly. The extract is concentrated to about three percent carrageenan before alcohol is added to precipitate it.

Chemically, it comprises repeating galactose units and 3,6-anhydrogalactose (3,6-AG), sulfated and non-sulfated, joined by alternating α (1-)-and β (1-4)-glycosidic linkages.(Figure2.4) Commercial carrageenans are available as stable sodium, potassium, and calcium salts or, most generally, as a mixture of these. The associated cations together with the conformation of the sugar units in the polymer chain determine the physical properties of the carrageenans. Carrageenans can be produced via a variety of process techniques; alcohol extraction, potassium chloride gel press or extracted with various alkalis. The process technique is important because it influences the gel characteristics. Its use in industrialized countries is well established and will rapidly spread into third world countries as their economies and demands for quality foods, cosmetics and industrial goods grow. (Chaplin 2008)

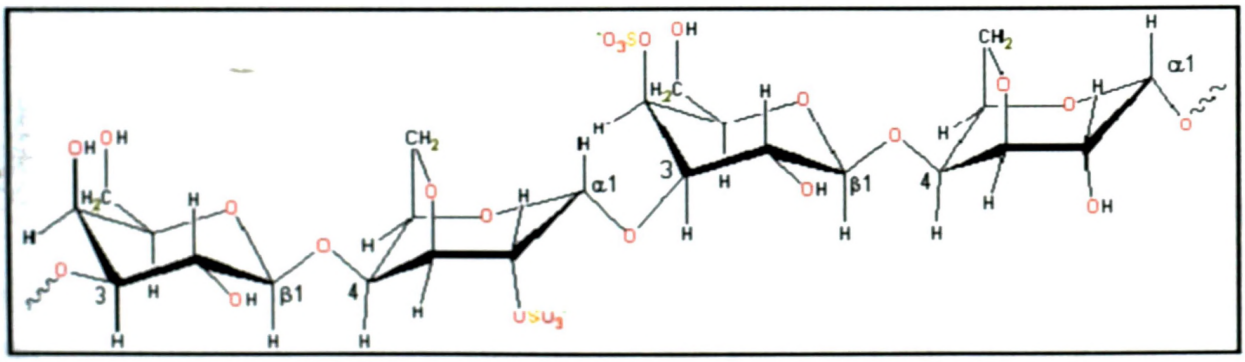


Figure 2.4 Carrageenan consists of alternating 3-linked-β-D-galactopyranose and 4-linked-α-D-galactopyranose units. (Source: Chaplin 2008)

2.7.1.1. Types of Carrageenan

There are three basic types of Carrageenan .They are kappa ,iota and lambda. Kappa carrageenan is the most commonly used type of carrageenan. Its most important properties are its high gel strength and strong interaction with milk proteins. About 70%

of the worlds carrageenan production is based on kappa carrageenan. Kappa carrageenan binds water to form strong, rigid gels. Potassium salts are essential in order to form this firm gel structure. As the level of potassium is increased, the resulting gel structure becomes tightly aggregated and may cause syneresis (moisture on the gel surface). (Chaplin 2008)

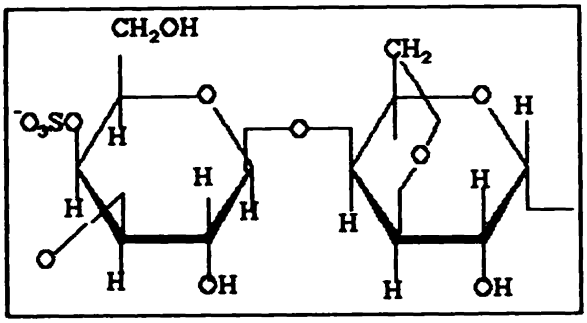


Figure 2:5 Kappa carrageenan (Source : MCPIcorperation 2009)

Iota carrageenan is is a type of carrageenan with a sulphate content intermediate between kappa and lambda carrageenan. Iota carrageenan also binds water, but forms a dry, elastic gel, especially in the presence of calcium salts. The divalent calcium ions help form bonds between the carrageenan molecules to form helices. The 2-sulfate group on the outside of the iota carrageenan molecule does not allow the helices to aggregate to the same extent as kappa carrageenan, but form additional bonds through calcium interactions. The gels are more elastic, dry and provide excellent freeze/thaw stability. (Chaplin 2008)

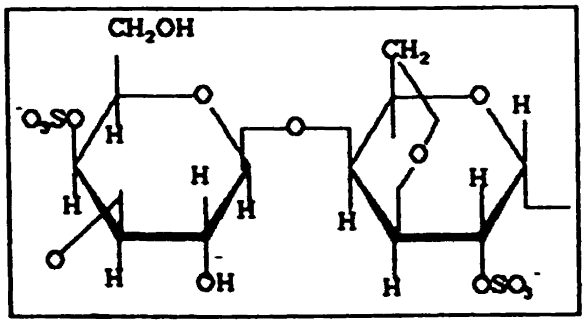


Figure 2.6 Iota carrageenan (Source: MCPIcorperation 2009)

Lambda carrageenan is highly sulfated and therefore less likely to form a gel structure. The ester sulfate distribution of lambda carrageenan is randomly distributed on the molecule. This prevents gelation and promotes viscous solutions. Lambda carrageenan is primarily used to thicken liquids and modify the texture of foods. The primary differences which influence the properties of kappa, iota and lambda carrageenan are the number and position of the ester sulfate groups on the repeating galactose units . Higher levels of ester sulfate lower the solubility temperature of the carrageenan and produce lower strength gels, or contribute to gel inhibition (lambda carrageenan). Commercially it is supplied as it is extracted from the seaweed which is as a kappa / lambda mixture

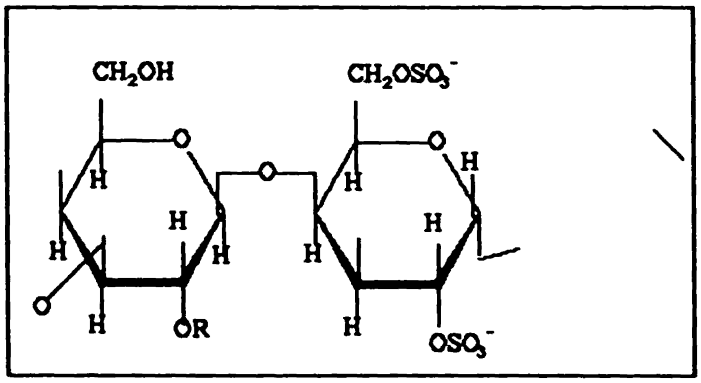


Figure 2.7. Lamda carrageenan (Source :MCPIcorperation 2009)

2.7.1.2. The Mechanisum of Carrageenan

Probably the best-known synergistic carrageenan interaction is that involving milk proteins. In these applications, the kappa carrageenan forms a weak gel in the aqueous phase and it interacts with positively charged amino acids in the proteins at the surface of the casein micelles. The specific kappa carrageenan-kappa casein interaction is shown diagrammatically(Figure2.8.).Carrageenan is a highly negatively charged macromolecule and has the ability to interact with any species carrying an opposite charge molecules with positively charged groups (e.g., proteins below the isoelectric point) will complex directly with carrageenan without the need for intervening cations Above the isoelectric point, cations are required to form an electrostatic bridge between the protein and carrageenan The details of the interaction depend critically on the stereochemistry of the

protein. The interaction can lead to the precipitation of the protein to the formation of a stable complex or gel (MCPIcorperation 2009)

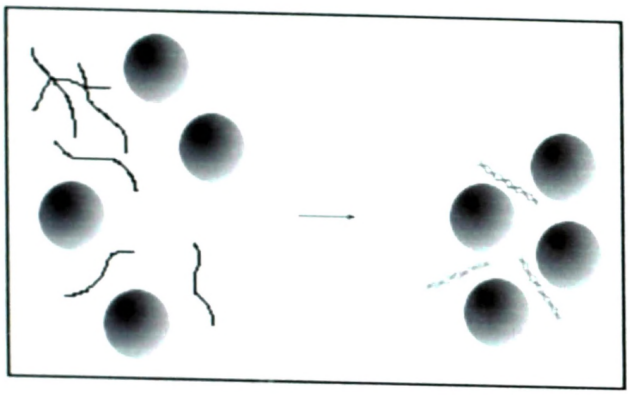


Figure 2.8. Carrageenan and milk proteins (Source CP Kelco 2008)

2.7.1.3. Functional Properties of Carrageenan

2.7.1.3.1. Gelling Agent

Depending on the combinations of different carrageenan fractions, a broad variety of gel textures can be obtained, from strong and brittle to very elastic. At temperatures higher than 60° C, Carrageenan exists in solution as a random coil which undergoes a double helix transition as the temperature decreases. Gels form when the double helices align to form quasi-crystalline regions.Requires the presence of cations for alignment.

Carrageenan solutions and gels are stable at neutral and slightly acid system. The combination of elevated temperature and acid conditions will produce hydrolysis of carrageenan resulting in a loss of viscosity and/or gel strength. In acid systems (pH < 3.7), it is advisable to add carrageenan at the latest possible stage during processing, and cool down the solution very quickly. Once the gel is formed, this effect is not observed

2.7.1.3.2. Water Holding Agent

Kappa and Iota carrageenans are excellent water holding agents due to their high capacity to bind water and form gels. This capacity allows them to retain the natural water of products, especially when subjected to thermal processing and to increase their processing yield.

2.7.1.3.3. Suspending Agent

Kappa at very low concentrations form an imperceptible weak gel in milk or water allowing solids to remain suspended in solution without providing much viscosity to the system.

2.7.1.3.4. Thickening Agent

Lambda carrageenan may act as thickening agent in cold and hot systems. Iota and Sodium kappa carrageenans are also widely used as thickening agents in products that undergo thermal processing.

2.7.1.3.5. Stabilizing Agent

Carrageenans are able to stabilize emulsions because of their high capacity to form matrixes and their strong electrostatic interaction. Due to the high specificity of Carrageenan, it is the only agent capable to stabilize without modifying the system texture.

2.7.1.3.6. Viscosity of Carrageenan Solutions

Viscosity is a measure of the amount of shearing stress or liquid resistance to flow by a fluid or semi fluid. For Carrageenan solutions, the measured viscosity is affected by factors such as temperature, presence of cations and degree of gum hydration. When the Carrageenan powder is dispersed in water at room temperature, the particles absorb water leading to swelling and increase in size as the particles start to hydrate. With heating, the hydrated molecules uncoil and tend to intertwine with adjacent particles forming a viscous solution.As the temperature increases,swelling increases and so does the measured viscosity as the particle becomes fully swollen. Further heating eventually leads to complete dissolution of Carrageenan which leads to a decrease in viscosity.

2.7.1.3.7. Interaction with Salts

The ability of Carrageenan to form a gel and the characteristics of the gel formed, is related to how closely the Carrageenan molecules can align to form a quasi-crystalline

network.The presence of ester sulfates tends to keep the molecules apart thus the need for cations to act as a bridge between two molecules.The functionality of Carrageenan is sensitive to both the type and concentration of cation.of the three types, *lambda* is the least salt sensitive (lambda is non gelling) and kappa the most. The physics behind is not well defined.Sodium and potassium salts of polyphosphates and citrates enhance solubility of Carrageenan in cold and hot solutions and reduce their viscosity due to divalent cations chelation.(MCPIcorperation 2009)

2.7.1.4. Stability of Carrageenan Powder

Carrageenan powders contain 8-10 % moisture, most of it bound to the molecule. It does not absorb moisture from the atmosphere or cause other ingredients to cake.The dry powder may be stored for a year or more under standard warehousing conditions without deterioration or loss in quality so long as it remains dry.Unlike other gums, carrageenan is insensitive to enzymes, especially cellulase.In both gel and solution form, food preservatives must be added to prevent bacterial contamination, which may cause fermentation and eventually, the degradation of the Carrageenan. However, aseptically packed products do not require .preservatives.Carrageenan is highly stable in boiling neutral or alkali solutions without loss in viscosity or gel potential.In acidic systems, Carrageenan solutions are susceptible to viscosity losses specially at high cooking temperature.In gel form, Carrageenan is stable even at low pH. (MCPIcorperation 2009)

2.7.1.5. Application of Carrageenan

2.7.1.5.1. Food Application

- Processed Meat - substitutes fat and serves as meat extender and binder; enhances juiciness;increases yield; prevents fat separation
- Processed Poultry - controls dehydration while frozen; enhances juiciness and increases yield
- Milk/Chocolate Milk Drink/Juice - stabilizes and improves viscosity

- Ice Cream - prevents large ice crystal formation; enhances excellent flavor release
- Flan/Dessert Gel/Confectionery - serves as gelling agent; enhances flavor release and excellent mouthfeel
- Bread/ Noodle/Pasta - increases yield and improves texture and mouthfeel
- Cakes/Pastries -substitutes butter and improves texture and mouthfeel
- Sauce/Salad Dressing - thickens and improves viscosity
- Beer/Wine/Vinegar - accelerates and improves clarity

2.7.1.5.2. Non Food Application

- Moist meat > serves as gelling and stabilizing agent
- Moist whole fish > serves as binder
- Culture Media > serves as gelling agent and stabilizer
- It apply to produce toothpaste and air fresheners

2.7.1.5.3. Pharmaceutical Application

Pharmaceutical application :Insoluble carrageenan- chitosan fibers can be spun with active pharmaceutical agents trapped within the fibers. The resulting systems, although water insoluble, will absorb considerable quantities of body fluids enabling wounds to be kept clean and dry speeding the healing process.

2.7.1.6. Safety Aspect of Carrageenan

The WHO (world health organization) has set an acceptable daily intake of carrageenan of " not specified" since the total daily intake (arising from it's use at level necessary to achieve the desire effect and from its background in food) was not consider to represent a hazard to health. The complex polysaccharide may be degraded a little in the acid environment of the stomac, but no enough to cause any harm.

In the UK food advisory committee has recommend that carrageenan should not be used as an additive for infants' formulation . ADI value is 75 mg/ kg body weight.

2.7.2. Sodium Citrates

Sodium salts of citric acid, a compound found in every living organism, as it is part of the key metabolic pathways in all body cells. Large concentrations are found in citrus fruits, kiwi, strawberries and many other fruits. Commercially prepared by fermentation of molasses with the mould *Aspergillus niger* .

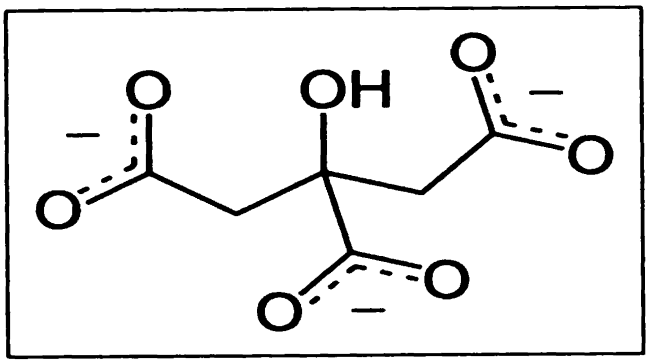


Figure 2.9. Citrate anione (Source: Wikipidia 2008)

2.7.2.1. Monosodium Citrate

Monosodium citrate has the chemical formula $NaH(C_3H_5O(COO)_3)$. Since it has two remaining open spots on the citrate anion, it is used as a relatively strong.(Wikipedia 2008)

2.7.2.2. Disodium Citrate

Disodium citrate is a citric acid sodium salt with the formula .$Na_2H(C_3H_5O(COO)_3)$. It is used as an ntioxident in food as well as to improve the effects of other antioxidants. It is also used as an acidity regulator and sequestrant. (Wikipedia 2008)

2.7.2.3. Trisodium Citrate

Trisodium citrate has the chemical formula of $Na_3C_3H_5O(COO)_3$. It possesses a saline, or mildly tart, flavor. Trisodium citrate is chiefly used as a food additive usually for flavor or as a preservative Trisodium citrate is employed as a flavoring agent in certain varieties of club soda.(Wikipedia 2008)

2.7.2.3. Application of Sodium Citrates

- Sodium citrate is used in ice cream to keep the fat globules from sticking together. Citrates and phosphates both have this property.
- It is also an anti-coagulant.
- As a buffering agent, sodium citrate helps maintain pH levels in soft drinks.
- As a sequestering agent, sodium citrate attaches to calcium ions in water, keeping them from interfering with detergents and soaps.

2.8. Chocolate liquid

The chocolate liquid formed in an intermediate stage is used in the confectionery trade as a covering for fruits, candies, or cookies, or the process may be continued and the resulting smooth mass of chocolate molded, cooled, and packaged as candy. It should be hard enough to snap when broken, have a mellow flow when melting, be free of gritty particles, and have a rich, dark color and an aromatic smell and flavor.

2.9. Basic operation

2.9.1. Pasteurization milk

Tested raw milk is to be processed it is first heated to about 72 °C for 15s in a plate heat exchanger. Plate heat exchanger in which milk is rapidly heated and rapidly cooled down to a certain temperature depending on the size of regenerative and cooling section .The milk is then clarified. After clarification milk is standardized. The process of adjusting the fat content of milk and cream to certain levels is known as standardization. Then ingredient of chocolate pasteurized milk are mixed and heated in to 45 °C. After heating, mixture is homogenized (150kg /cm^2) and pasteurized (75-80 °C/15s). Then the mixture is chilled (16 °C). Finished mixture fills in to tetra pack and store at 4 °C. (Figure 2.10)

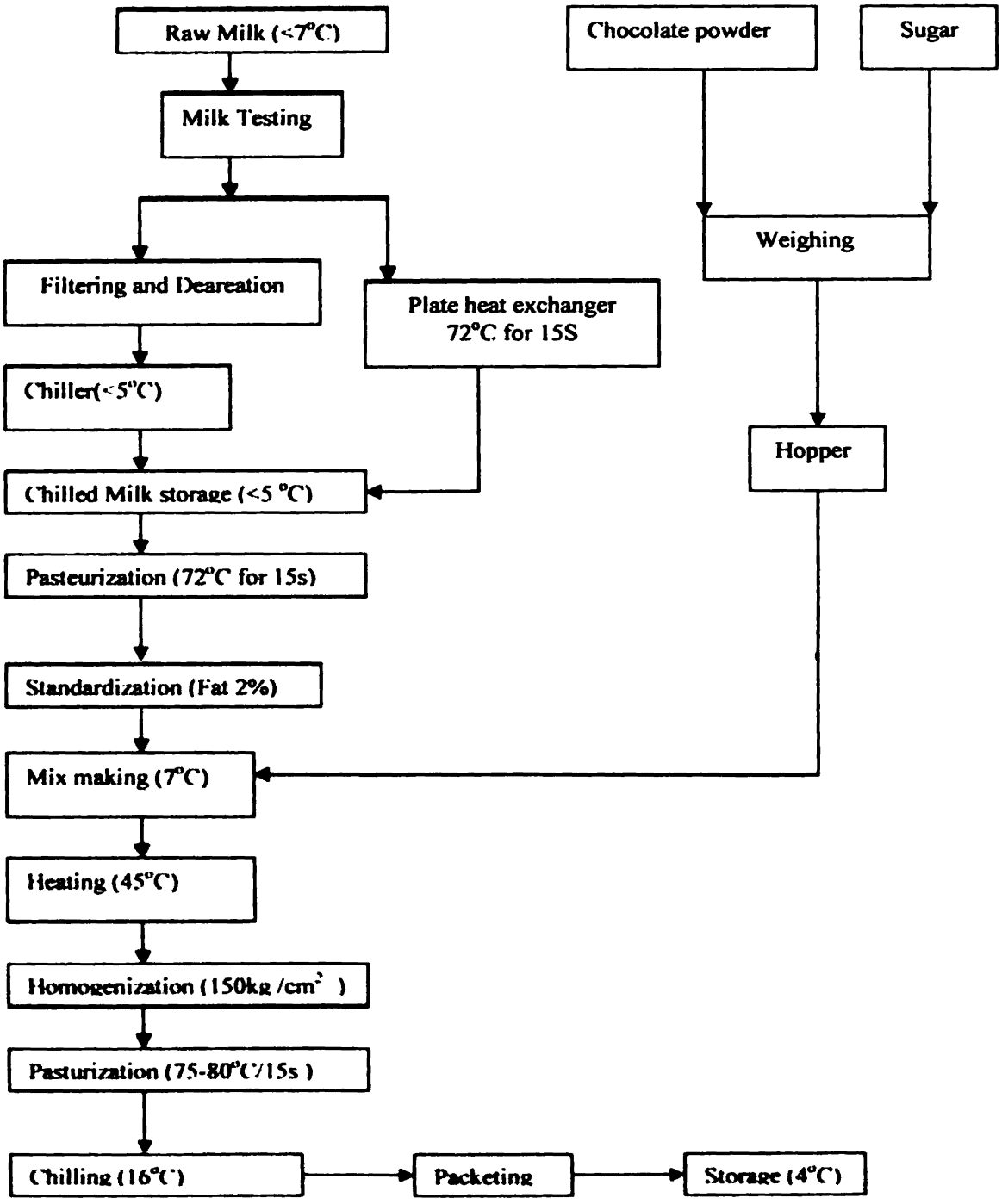


Figure 2.10. Process flow diagram of chocolate flavored pasteurized milk

2.9.2. Sterilized milk

Tested raw milk is to be processed it is first heated to about 72 °C for 15s in a plate heat exchanger. Plate heat exchanger in which milk is rapidly heated and rapidly cooled down to a certain temperature depending on the size of regenerative and cooling section .The milk is then clarified. After clarification milk is standardized. The process of adjusting the fat content of milk and cream to certain levels is known as standardization.

The purpose is to produce product with an uniform compositional quality.The level to which the fat content of milk can be adjusted are the minimum fat contents established by the relevant government.Then ingredient of chocolate in – container sterilized milk are mixed. After mixing the ingredients, mixture is presterilized.

Then this mixture is homogenized. The purpose of the homogenization of the whole milk is get to the stable emulsion, which will not be to form a cream plug layer and also makes it possible to reduce the fat content without affecting the flavour. Homogenized mixture is heated to 60 °C and filled in to clean glass bottles and capped. The filled, closed and labeled bottles are then led by a chain conveyer into a tower. The temperature is normally 115- 120 °C with towers at 10 m in height. After the sterilization in tower, bottles are stored in room temperature. (Figure 2.11.)

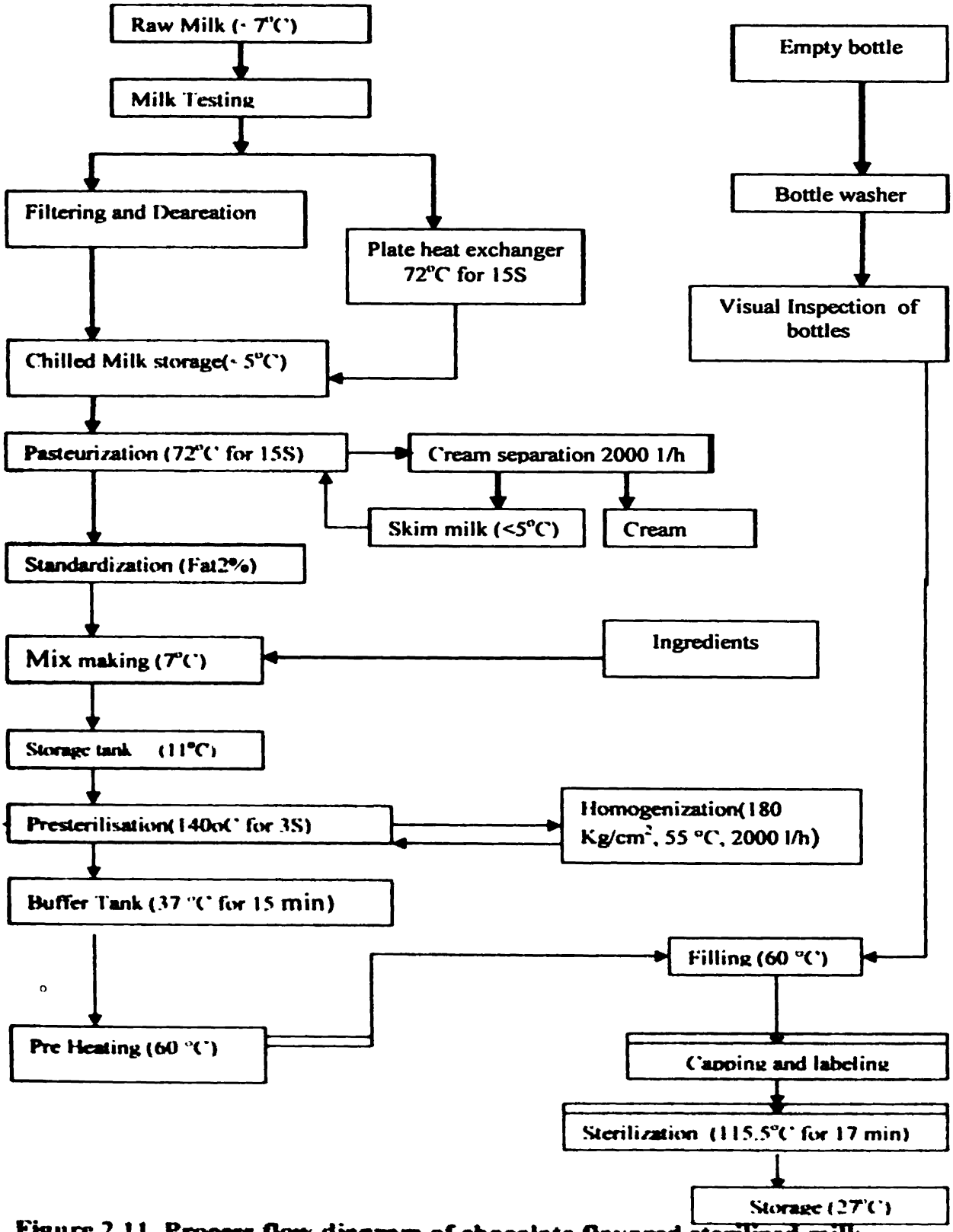


Figure 2.11. Process flow diagram of chocolate flavored sterilized milk

2.10. Determination of product specification

2.10.1. Determination of Fat

The Gerber Method is a primary and historic chemical test to determine the fat content of milk and other substances. Milk fat is separated from proteins by adding sulphuric acid. The separation is facilitated by using amyl alcohol and centrifugation. The fat content is read directly via a special calibrated butyrometer. Gerber developed specialized butyrometers. (Atherton and Newlander 1987)

2.10.2. Determination of Acidity

It is a principle chemistry that an alkali will neutralize an acid that is when these two chemicals are mixed together products will be formed that are acid neither acid nor bases. The principal is made use of in testing milk for acid.By measuring the amount of alkali of a given strength that it takes neutralize the acid in a sample of milk.The process of determining the amount of acid in a sample is called titration.Tenth normal sodium hydroxice is the alkali used and phenolphalein is the indictor for showing when all the acid has been neutralized. Phenolphalein is colouless in an acid medium , but turns pink in an alkali solution. (Atherton and Newlander 1987)

2.11. Microbiological Test

The keeping quality of the pasteurized milk depends on the effect of heating, recontamination and temperature of cold storage. The effect of heating depends on the number of bacteria able to survive low pasteurization (72 °C/15s) of which the most common.

a .Streptococci (Enterococci)

b. Lactobacilli (certain species)

c.Bacteria spores (bacillus, clostridium)

Coli,Streptococci and Pseudomonas are bacteria which are found in recontaminated milk. The rate of multiplication of these bacteria in milk depends on the temperature of storage. At low temperature 0 °C to 5 °C a slow multiplication of the psycrophillic bacteria takes place. The bacteria which are pasteurization surviving, are to a great extent the proteolytic and lipolytic bacteria, and the milk becomes bitter and rotten. At higher storage temperature 15 °C to 25 °C growth of lactic acid bacteria predominates and the milk gets an impure and sour taste. (FAO 1975)

2.11.1. Metheylene Blue Reduction Test

The methylene blue reduction test is based on the fact that the color imparted to milk by the addition of a dye such as methylene blue will disappear more or less quickly. The removal of the oxygen from milk and the formation of reducing substances during bacterial metabolism cause the color to disappear. The agencies responsible for the oxygen consumption are the bacteria. Though certain species of bacteria have considerably more influence than others, it is generally assumed that the greater the number of bacteria in milk, the quicker will the oxygen be consumed, and in turn the sooner will the color disappear. Thus, the time of reduction is taken as a measure of the number of organisms in milk although actually it is likely that it is more truly a measure of the total metabolic reactions proceeding at the cell surface of the bacteria.

The methylene blue reduction test has lost much of its popularity because of its low correlation with other bacterial procedures. This is true particularly in those samples which show extensive multiplication of the psychrotropic species.(Atherton and Newlander 1987)

Table2.5. Suggested classification is listed. (Source: Atherton and Newlander1987)

Class 1.	Excellent	not decolorized in 8 hours
Class 2.	Good	decolorized in less than 8 hours but not less than 6 hours
Class 3.	Fair	decolorized in less than 6 hours but not less than 2 hours
Class 4	Poor	decolorized in less than 2 hours

Many factors affect the methylene blue reduction and steps of operation should be uniform. Since the oxygen content must be used up before the color disappears, any manipulation that increases the oxygen affects the test. Cold milk holds more than warm milk; pouring milk back and forth from one container to another increase the amount, and milking time much oxygen may be absorbed. The kind of organisms affects the rate of reduction. The coli forms appear to be the most rapidly reducing organisms, closely followed *by Streptococococcus lactis*, some of the faecal *Streptococci*,And certain micrococci. Thermoduric and psychrotrophic bacteria reduce methylene blue very slowly if at all. A large number of leucocytes affect the reduction time materially.Light hastens reduction and therefore the tests should kept covered.The concentration of the dye should be uniform as an increased concentration of the dye should be uniform as an increased concentration lengthens the time of reduction. Increasing the incubation temperature augments the activity of the bacteria and therefore shortens the reduction time. The creaming of the test samples causes a number of organisms to be removed from the body of the milk and brought to the surface with the rising fat. This factor causes variation in the reduction time, since the bacteria are not evenly distributed. The accuracy of the test is increased, reduction time shortened and decolonization more uniform if the samples are periodically inverted during incubation. (Atherton and Newlander 1987)

2.11.2. Total Colony Count Test

The estimation of the number of bacteria in milk by the plating procedure consists in mixing a definite volume of milk with melted agar to solidify and then counting the colonies that appear upon incubation .A colony consists of the progeny of one cell or group of cells, and results are reported as so many colonies per milliliter of milk.

2.12. Sensory Evaluation

Sensory analysis is the identification; scientific measurements, analysis and interpretation of the properties (attributes) of a product as the y are perceived through the five senses of sight, smell, taste, touch and hearing Sensory analysis answers questions of quality under three main headings-discrimination, description and preference. In discrimination questions aim to find out whether or not a difference exist between two or more products.

In description, questions aim to describe and measure any difference that is found to exist between products. In preference, hedonic questions aim to identify liking or acceptability (Roland *et a.* 2000).

The test may be combined with scoring, in which samples are evaluated on a predetermined numerical basis covering a single quality factor or a combination of quality factors (Cloninger et al.1976) Scales may include any number of steps, depending on the degree of precision possible in making judgment.

CHAPTER 03

MATERIALS AND METHODOLOGY

3.1. Materials

3.1.1. Materials

Three 250ml in - container sterilized milk bottles and pasteurized milk packets (produced by MILCO factory)

Pasteurized milk

Carrageenan(E407)

Milk (Fat 2.0%)

Sugar

Cocoa powder(10-12% Fat)

Chocolate liquid

Sodium citrate (E331)

3.1.2. Apparatus

Analytical balance (Weighing up to 220.0000g, Accuracy .0001)

Desiccator

Oven (maintained at 105 ±1°C)

Water bath (maintained at 65°C)

Centrifuger (1100rpmfor 5.min)

Butyrometer

Moisture dishes made of Aluminum

Labouratory glasswares

Beakers

Thermometer (±1^{0}C)

Pipettes

Petri dishes

Test tubes

Measuring cylinder

Empty 25 glass sterilized bottles

Auto clave

Rubber stoppers

Incubator(35±1^{0}C for72±hrs)

Colony counter

3.1.3. Reagents

Gerber Sulphuric acid (90-91% w/v)

Amyl alcohol(70% w/v)

0.1M NaOH

1% phenolphthalein

Methylene Blue

Agar

3.2. Determination of the Amount of Sediment

Methodology:

At first, present sediment was analyzed. Three bottles were taken before two days from the production and let them to settle the sediment. Pre-dried three Petri dishes were weighted by using analytical balance. After removing Supernatant, the remaining sediment of three samples was weighted quickly into the each petri dish. Then they were kept in oven at 105±1^{0}C for 4 hours.Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987) Then weights of sediments were recorded on weight percent basis as follows:

$$\text{Sediment percentage \%} = \frac{(\text{Initial Weight} - \text{Final Weight})}{\text{Initial Weight}} \times 100\%$$

Equation. (1)

3.2.1. Determination of Fat

Methodology:

About 10ml of Gerber sulphuric was added to the butyrometer. 10.94 ml of sterilized milk was pippetted put in to the butyrometer. Then 1ml of amyl alcohol and sufficient warm distilled water were added so that the butyrometer was filled to the shoulder below the neck. The tube was closed with stopper and mixed the content thoroughly .Then tube was centrifuged at 1100 rpm for about 5 minutes. Finally tube was returned to the water bath and after 5 minutes percentge of fat was read directly on the scale.This was triplicated (Atherton and Newlander 1987)

3.2.2. Determination of Total Solids

Methodology:

Pre-dried 25 aluminum dishes were weighted to the nearest 0.1 mg on an analytical balance. Then 2 g of fragmented chocolate milk was weighted quickly into the each aluminum dishes and kept at 105±1^{0}C for 4 hours. Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987)

Then total solids and moisture contents were recorded on weight percent basis as follows:

$$\text{Total Solids\%} = \frac{(\text{Initial Weight} - \text{Final Weight})}{\text{Initial weight}} \times 100\%$$

Equation (2)

Moisture 100 - Total Solids%

Equation (3)

3.2.3. Determination of Acidity

Methodology:

9 ml of sterilized milk of each samples were titrated with 0.1M NaOH, using 1ml phenolphthalein as an indicator and acidity was calculated as lactic. (Atherton and Newlander 1987)

[1ml of 0.1M · 0.09 g lactic acid]

3.3. Selection of best stabilizer appropriate amount

3.3.1. Test for Carrageenan

Methodology

The amount of Carrageenan was seleced as follows,

Table 3.1. Added amount of Carrageenan

Carrageenan (%)	0.01	0.02	0.03	0.04	0.05
Carrageenan weight (g)	0.0265	0.0530	0.0795	0.106	0.1325

Cocoa powder, milk and Carrageenan were measured using analytical balance as following table,

Table 3.2 Tested formulas using Carrageenan

Ingredient	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Milk	230ml	230ml	230ml	230ml	230ml
Sugar	17.85g	17.85g	17.85g	17.85g	17.85g
Cocoa powder	2.0917g	2.0917g	2.0917g	2.0917g	2.0917g
Carrageenan	0.0265g	0.0530g	0. 0795g	0.1060g	0.1325g

100 ml of milk, sugar ,cocoa powder and Carrageenan were mixed to prepare chocolate syrup .The mixture was heated using water bath at 70 °C for 20 minutes until the ingredients were well dissolved. Then 130 ml of milk was added to the chocolate syrup and pasteurized in water bathe while stirring.After the mixture was taken out and filled 250 ml remaining 1.5 inches head space for each pre sterilized 25 glass bottles and capped. Then filled bottles were put in to the autoclave. The autoclave was then heated with steam under a pressure corresponding to the required temperature (110-120°C for 30-45 minutes).After 45 minutes processing time, the steam was vented to atmosphere. The auto clave was opened and the crates of the bottles were removed for natural air.

Pre-dried 25 aluminum dishes were weighted to the nearest 0.1 mg on an analytical balance. Then 2 g of fragmented chocolate milk was weighted quickly into the each aluminum dishes and kept at 105±1°C for 4 hours. Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987)Then total solids and moisture contents were recorded on weight percent basis as follows

Then fat (%), total solids (%), and acidity were measured according to procedures mentioned in 3.2.1, 3.2.2 and 3.2.3

3.3.2. Test for Sodium citrate

Methodology:

Amount of Sodium citrate was added as follows,

Table 3.3. Amount of Sodium citrate

Sodium citrate (%)	0.01	0.03	0.05	0.08	1.00
Sodium citrate weight (g)	0.0265	0.0795	0.1325	0.212	2.65

Ingredients were measured according to the following table.

Table 3.4 Tested formulas using Sodium citrate

Ingredient	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5
Milk	230ml	230ml	230ml	230ml	230ml
Sugar	17.85g	17.85g	17.85g	17.85g	17.85g
Cocoa powder	2.0917g	2.0917g	2.0917g	2.0917g	2.0917g
Sodium citrate	0.0265g	0. 0795g	0.1325g	0.2120g	2.650g

100 ml of milk, sugar ,cocoa powder and Sodium citrate were mixed to prepare chocolate syrup .The mixture was heated using water bath at 70 °C for 20 minutes until the

ingredients were well dissolved. Then 130 ml of milk was added to the chocolate syrup and pasteurized in water bathe while stirring.After the mixture was taken out and filled 250 ml remaining 1.5 inches head space for each pre sterilized 25 glass bottles and capped. Then filled bottles were put in to the autoclave. The autoclave was then heated with steam under a pressure corresponding to the required temperature (110-120°C for 30-45 minutes).After 45 minutes processing time; the steam was vented to atmosphere. The auto clave was opened and the crates of the bottles were removed for natural air.

The bottles were allowed for two days to settle sediment.

Pre-dried 25 Petri dishes were weighted. After pippetting out supernatant, the remaining sediment of 25samples were weighted quickly into the each aluminum dishes and kept at 105±1°C for 4 hours. Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987) Then weights of sediments were recorded on weight percent basis as follows:

Calculation was done on Equation (1)

Then fat (%) , total solids(%), and acidity were measured according to mentioned procedures in 3.2.1, 3.2.2 and 3.2.3.

3.3.3. Test for Combination of Sodium citrate and Carrageenan

Methodology:

Amount of Carrageenan and Sodium citrate combination was selected to prepare 1,2,3,4and 5by changing the level of Carrageenan as 50%, 40%, 30% ,20%,10%and as Sodium citrate 50%, 60%, 70%,80%,90% for each sample respectively.

Table 3.5. Added amounts of Carragenan and Sodium citrate

Stabilizer	Sample 1	Sample2	Sample3	Sample4	Sample5
Carragenan (%)	50	40	30	20	10
Carragenan weight(g)	0.795	0.636	0.477	0.318	0.159
Sodium citrate (%)	50	60	70	80	90
Sodium citrate weight(g)	0.795	0.954	1.113	1.272	1.431

Cocoa powder, milk, Carrageenan and Sodium citrate were measured using analytical balance as following table.

Table 3.6 Tested formulas using combination of Carrageenan and Sodium citrate

Ingredient	Sample 1	Sample 2	Sample3	Sample4	Sample5
Milk	230ml	230ml	230ml	230ml	230ml
Sugar	17.85g	17.85g	17.85g	17.85g	17.85g
Cocoa powder	2.0917g	2.0917g	2.0917g	2.0917g	2.0917g
Carrageenan	0.795g	0.636g	0.477g	0.318g	0.159g
Sodium citrate	0.795g	0.954gg	1.113g	1.272g	1.431g

100 ml of milk, sugar ,cocoa powder ,Carrageenan and Sodium citrate were mixed to prepare chocolate syrup .The mixture was heated using water bath at 70 °C for 20 minutes until the ingredients were well dissolved. Then 130 ml of milk was added to the chocolate syrup and pasteurized in water bathe while stirring.

After the mixture was taken out and filled 250 ml remaining 1.5 inches head space for each pre sterilized 25 glass bottles and capped. Then filled bottles were put in to the autoclave. The autoclave was then heated with steam under a pressure corresponding to the required temperature (110-120°C for 30-45 minutes).After 45 minutes processing time; the steam was vented to atmosphere. The auto clave was opened and the crates of the bottles were removed for natural air.

The bottles were allowed for two days to settle sediment. After two days, Pre-dried 25 Petri dishes were weighted. After pippetting out supernatant, the remaining sediment of 25samples were weighted quickly into the each aluminum dishes and kept at 105±1°C for 4 hours. Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987)Then weights of sediments were recorded on weight percent basis as follows:

Calculation was done on Equation (1).Then fat % , total solids, and acidity were measured according to mentioned procedures in 3.2.1, 3.2.2 and 3.2.3.

3.4. Selection of the Best Formula (In –container sterilized milk)

Methodology:

Three in-container sterilized milk samples were prepared by changing the level of chocolate liquid as 90%, 60%, 30% and cocoa powder as 10%, 40%, 70%, for each sample respectively and level of Carrageenan was 0.01% (Finally estimated amount of Carrageenan)for each, while keeping the other ingredients constant.

Table 3.7 The changing level of cocoa powder and chocolate liquid (In- container sterilized milk)

Ingredients	Percent (%)	Weight(g)
Cocoa powder	90	0.11747
	60	0.83668
	30	1.46419
Chocolate liquid	10	1.88253
	40	1.25502
	70	0.62751

Cocoa powder, milk, Carrageenan and were measured using analytical balance as following table.

Table 3.8. Tested formulas (In- container sterilized milk)

Ingredient	436	542	678
Milk	230ml	230ml	230ml
Sugar	17.85g	17.85g	17.85g
Cocoa powder	0.11747g	0.83668g	1.46419g
Chocolate liquid	1.88253g	1.25502g	0.62751g
Carrageenan	0.0265g	0.0265g	0.0265g

100 ml of milk, sugar ,cocoa powder , chocolate liquid and Carrageenan were mixed to prepare chocolate syrup .The mixture was heated using water bath at 70 °C for 20 minutes until the ingredients were well dissolved. Then 130 ml of milk was added to the chocolate syrup and pasteurized in water bathe while stirring.After the mixture was taken out and filled 250 ml remaining 1.5 inches head space for each pre sterilized 25 glass bottles and capped. Then filled bottles were put in to the autoclave. The autoclave was then heated with steam under a pressure corresponding to the required temperature (110-120°C for 30-45 minutes).After 45 minutes processing time, the steam was vented to atmosphere. The auto clave was opened and the crates of the bottles were removed for natural air.

The bottles were allowed for two days to settle sediment.

Pre-dried 25 Petri dishes were weighted. After pippetting out supernatant, the remaining sediment of 25samples were weighted quickly into the each aluminum dishes and kept at 105±1°C for 4 hours. Thereafter samples were cooled in a desiccator and total dry weight was determined. Again, samples were kept for an additional 20 minutes inside the oven and reweighed. This was repeated several times until constant weight was attained. Once the difference between the two consecutive readings after the additional drying

period was less than 1 mg, it was recorded as the final reading. (Atherton and Newlander 1987)Then weights of sediments were recorded on weight percent basis as follows:

Calculation was done on Equation(1)

Then fat % , total solids(%), and acidity were measured according to mentioned procedures in 3.2.1, 3.2.2 and 3.2.3.

3.5. Selection of the Best Formula (Pasteurized Milk)

Methodology:

Three pasteurized milk samples were prepared by changing the level of chocolate liquid as 20%, 40%, 60% and cocoa powder as 80%, 60%, 40% while keeping the other ingredients constant

Table3.9. The changing level of cocoa powder and chocolate liquid (In pasteurized milk)

Ingredients	Percent(%)	Weight(g)
Cocoa powder	80	0.66820
	60	0.50120
	40	0.33408
Chocolate liquid	20	0.16700
	40	0.33408
	60	0.50112

Table3.10. Tested formulas(In pasteurized milk)

Ingredients	357	432	524
Milk	168 ml	168ml	168ml
Sugar	11.7 ml	11.7ml	11.7 ml
Chocolate powder	0.6682g	0.50120g	0.33408g
Chocolate liquid	0.16700g	0.33408g	0.50112g

The measured milk, sugar ,cocoa powder and chocolate liquid were mixed .The mixture was heated using water bath at 80°C for 20 minutes until the mixed ingredient were dissolved well. After the mixture was taken out and put in ice bathe at 4°C and store in refrigerator.

Then fat (%) , total solids(%), and acidity were measured according to mentioned procedures in 3.2.1, 3.2.2 and 3.2.3.

3.5.1. Methylene Blue Reduction Test for Formulas

All glassware and rubber stoppers were sterilized in boiling water. 1 ml of the methylene blue solution was measured into a test tube. 10 ml of pasturized milk was added. Stopper was put in to the test tube. Tubes were placed in the water bath immediately for a more convenient time of incubation. When ready to perform the test, the temperature of the samples should be brought to 35°C within 10 minutes.

When temperature reaches 36°C, slowly invert tubes a few times to assure uniform creaming. Tubes were not shaken. This time was recorded as the beginning of the incubation period. They were covered to prevent exposure to light.

Samples for decolonization were checked after 30 minutes of incubation. Subsequent readings were obtained at hourly intervals thereafter. After each reading, decolorized tubes were removed and then gently one complete inversion of remaining tubes were made. Reduction time was recorded in whole hours between last inversion and decolonization.

3.5.2 Total Colony Count by Using Pour plate method

Methodology:

The distribution serial 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4} solutions were prepared with15ml of the prepared standard 15ml of the prepared standard method agar medium was pour into the Petri dish as 45 ± 0.5°C .Then 1ml from each sample of serial dilution was pipetted out and introduce aseptically into the serial dilution pleats .It were label as a 1ml was pipetted out and introduce aseptically into sterilized plates. It ware label as a 10^{-1} 10^{-2},10^{-3},10^{-4}1ml

was pipetted out from the original sample and it was introduced into Petri dish with 15ml of prepared standard method agar medium. It was labored then lid was closed immediately and shaken gently for even distribution of media on plate.

After that plates were kept invented on a clean horizontal surface for few minutes to solidify the nutrient agar then dishes were surface and incubate at 35 ± 1°C for 72 ± 3hrs after the specified period of incubation, colonies were counted in each Petridis using the colony counter (SLS 516 part I 1991)

3.6. Sensory Evaluation

.The sensory evaluation for three samples of pasteurized and three samples of sterilized was done by 30 untrained panelists in MILCO factory in Digana. Sensory evaluation was carried out under condition which avoids influences of external forces. Samples of sterilized milk and pasteurized milk were coded with 3 digit random numbers. These numbers were 436, 542 and 678 and 357, 432, and 524.Samples representing each ingredient combination were placed before the panelist alone with the ballot paper. Level of preference for each sensory attribute (taste, colour, smell, sedimentation and overall acceptability) in 3 samples of in container sterilized milk was recorded according to the 7 point hedonic scale. Level of preference for each sensory attribute (taste, colour, smell, appearance and overall acceptability) in 3 samples of pasteurized milk was recorded according to the 7 point hedonic scale. Obtained result analyzed by Minitab 14.1 statistical analysis software with use of Friedman test at 5% significant level.

CHAPTER 04

RESULTS AND DISCUSSION

4.1. Results for Sediment before Modifying

Initial product of in-container sterilized milk was used to estimate present sediment. Amount of three samples, which were randomly selected, was estimated using oven - drying method. The average reading of Initial sediment percent observed was 81%.

4.2. The Product Specification (In –container sterilized milk)

The product specification was estimated as follows.

Table4.1. product specification of initial in container sterilized milk

Specification	Amount
Fat percent	2.00%
Total solids	17.5%
Acidity	0.15

Analyzed results initial products of the in-container sterilized milk were 2.0%of fat, 0.15 acidity, and 17.5% of total solids.

4.3. Test results for Carrageenan

For the different the levels of Carragenan as 0.01%, 0.02%, 0.03%, 0.04% and 0.05%for the sample1, 2, 3, 4and 5 respectively, sediment percents were finalized in the table below.

Table4.2. Tested sediment percentage for Carrageenan.

Samples	Sediment %
1	16 0%
2	12.5%
3	10.0%
4	No sediment
5	No sediment

Sediment percentageof them were16.0%.12.5%and 10.0%for sample1,2 and 3respectivly.

Sample 4 and 5 were not observed sediment.In sample 3, 4, and 5 can be a supernatant water layer was observed as on the top of the bottles. And at the bottom of the same bottles a solid gelly could be observed. The sample 1and 2 were comprised of much better colour and appearance to the naked eye. Based on the results observed, high dosages of Carrageenan cause the milk transferred in to a rigid solid like form and a gelly like form The stabilizer should be used at a minimum level without harming the nature of in container sterilized milk to avoid sedimentation.

If too high amount of stabilizer used, it caused to increase viscosity in the in –container sterilized milk. To hold cocoa powder particles in suspension, relatively high viscosity is required. A stabilizer such as kappa-carrageenan can be used to react with milk proteins and cocoa particles to form a three-dimensional network that holds the particles in suspension. In chocolate milks, this low level of Carrageenan is able to prevent separation and generate a stabilizing network, which maintains the cocoa particles in suspension Three types of Carrageenan mixture available in market.Hence this mixture used to carry out the rasceh.The obtained amounts of Carrageenan 0.05%.0.08% and1.00% were not applicable due to the observations to reduce sediment due to the above observations.

The product specification (Appendix (xiii)) were observed as follows.

Table 4.3. Tested product specification for Carrageenan

Samples	Acidity	Total Solid (%)	Fat (%)
1	0.150	17.6	2.0
2	0.155	17.55	2.0
3	0.160	17.50	2.0
4	2.000	17.20	2.0
5	2.100	17.24	2.0

4.4. Test results for Sodium citrate

For the different the levels of Sodium Citrate 0.01%,0.03%,0.05%,0.08% and1.00%for the sample1,2,3,4and 5respectively sediment percents were finalized in the table below.

Table4.4. Tested sediment percentage for Sodium Citrate.

Samples	Sediment %
1	24.0
2	14.5
3	No sediment
4	No sediment
5	No sediment

In sample 3, 4, and 5 can be a supernatant water layer was observed as on the top of the bottles. And at the bottom of the same bottles a solid gelly could be observed. They colour also very dark brown The sample 1and 2 were comprised of much better colour and appearance to the naked eye. Based on the results observed, high dosages of sodium citrate cause the milk transferred in to a rigid solid like form and a gelly like form The stabilizer should be used at a minimum level without harming the nature of in container sterilized milk to avoid sedimentation. Based on the results observed, high dosages of Sodium citrates cause the milk transferred in to a rigid solid like form and a gelly like

form The stabilizer should be used at a minimum level without harming the nature of in container sterilized milk to avoid sedimentation Also a high amount of sodium citrate made the milk more bitter taste.

The product specification(Appendix(xiii)) were observed as follows.

Table4.5. Tested product specification for Sodium citrate

Samples	Acidity	Total Solid (%)	Fat (%)
1	0.15	17.5	2.0
2	0.16	17.6	2.0
3	0.18	17.4	2.0
4	2.25	17.3	2.0
5	2.40	16.0	2.0

4.5. Test Results for the Combination Sodium citrate and Carrageenan

The sample1.2.3.4and5 which were changing the level of Carrageenan as 50%. 40%. 30% .20%.10%and as Sodium citrate 50%. 60%. 70%.80%.90% for each sample respectively sediment percents were finalized in the table below.

Table4.6 Tested sediment for the combination of Sodium citrate and Carrageenan

Samples	Sediment %
1	31.5
2	20.0
3	13.2
4	No sediment
5	No sediment

The sample 5 was observed a supernatant water layer as on the top of the bottles. Sample 3 was good in colour including a little sediment amount. Sample 1and 2 had t more sediment than sample 3.The product specification were observed as follows, Based on the results observed, high dosages of the combination of Carrageenan and Sodium citrates cause the milk transferred in to a rigid solid like form and a gelly like form The stabilizer should be used at a minimum level without harming the nature of in –container sterilized milk to avoid sedimentation

Table4.7. Tested product specification (Appendix (xiii)) for the combination of Sodium citrate and Carrageenan

Samples	Acidity	Total solid	Fat (%)
1	0.160	17.60	2.0
2	0.165	17.55	2.1
3	0.170	17.40	2.1
4	0.190	17.50	2.1
5	2.100	17.30	2.1

4.6. Tested sediment for the samples 436,542and 678

Sediment% of each sample estimated were given in the table below.

Table4.8 Tested sediment percentage for samples 436,542 and 678

Sample code	Sediment %
436	7.0
542	10.5
678	16.0

This Study of specification for the above two products were carried out ,to determine the quality . This study of specification was important and useful to know about limitation which are related to enhance organoleptic qualities. Sediment is an important organoleptic quality that can be visualized to the naked eye.

4.7. Results of Sensory Evaluation for in Container Sterilized Milk

After statistically analyses of the result of the sensory evaluation test, following results were obtained for each characteristic features of the sample.The result of effect on the taste of the in container sterilized milk is showing the highest rank for the product ,which contain chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P= 0.023of the test is less than minimum probability P=0.05. According to data sample chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for taste. Therefore this sample (code no 678) come under the category of like very much according to the 7-point hedonic scale

(Appendix (iii))

Table 4.9. Result of sensory evaluation test on taste in -container sterilized milk

Sample code	N	Median	Sum of rank
436	30	3.3333	49.0
542	30	4.0000	61.5
678	30	4.6667	69.5

The result of effect on the colour of the in -container sterilized milk is showing the highest rank for the product ,which contain chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P 0.003of the test is less than minimum probability P 0.05. According to data sample chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for colour. Therefore this sample (code no 678)come under the category of like very much according to the 7-point hedonic scale(Appendix(iv))

Table 4.10. Result of sensory evaluation test on colour in -container sterilized milk

Sample code	N	Median	Sum of rank
436	30	3.3333	50.5
542	30	3.6667	55.5
678	30	4.5000	74.0

The result of effect on the smell of the in –container sterilized milk is showing the highest rank for the product ,which contain chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant in the formula .According to data analysis there is a significant difference between the 3 samples. since probability value P= 0.027of the test is less than minimum probability P=0.05. According to data sample chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for smell. Therefore this sample (code no 678)come under the category of like very much according to the 7-point hedonic scale (Appendix(v))

Table 4.11. Result of sensory evaluation test on smell in -container sterilized milk

Sample code	N	Median	Sum of rank
436	30	4.1667	50.0
542	30	4.8333	60.0
678	30	5.5000	70.0

The result of effect on the sedimentation of the in container sterilized milk is showing the highest rank for the product ,which contain chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant in the formula .According to data analysis there is a significant difference between the 3 samples. since probability value P 0.008of the test is less than minimum probability P 0.05. According to data sample chocolate liquid 30% and cocoa powder 70%,with a

level of Carrageenan at 0.01% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for sedimentation. Therefore this sample (code no 678)come under the category of like very much according to the 7-point hedonic scale (Appendix(vi))

Table 4.12. Result of sensory evaluation test on sedimentation in -container sterilized milk

Sample code	N	Median	Sum of rank
436	30	4.000	51.5
542	30	5.000	55.5
678	30	6.000	73.0

The result of effect on the overall acceptability of the in –container sterilized milk is showing the highest rank for the product ,which contain chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P= 0.000 of the test is less than minimum probability P=0.05. According to data sample chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for overall acceptability . Therefore this sample (code no 678)come under the category of like very much according to the 7-point hedonic scale (Appendix(vii))

Table 4.13. Result of sensory evaluation test on overall acceptability in -container sterilized milk

Sample code	N	Median	Sum of rank
436	30	5.000	41.0
542	30	6.000	59.5
678	30	7.000	79.5

4.8. Results of Sensory Evaluation for Pasteurized Milk

The result of effect on the taste of the pasteurized milk is showing the highest rank for the product ,which sample containing 60% chocolate liquid and 40% cocoa powder in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P= 0.000of the test is less than minimum probability P=0.05. According to data sample chocolate liquid 60% and cocoa powder 40% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for taste. Therefore this sample (code no 524) come under the category of like very much according to the 7-point hedonic scale (Appendix (viii))

Table 4.14 Result of sensory evaluation test on taste in pasteurized milk

Sample code	N	Median	Sum of rank
357	30	3.000	43.5
432	30	4.000	62.5
524	30	5.000	74.0

The result of effect on the colour of the pasteurized milk is showing the highest rank for the product ,which sample containing 60% chocolate liquid and 40% cocoa powder in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P 0.000of the test is less than minimum probability P 0.05. According to data sample chocolate liquid 60% and cocoa powder 40% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for colour. Therefore this sample (code no 524)come under the category of like very much according to the 7-point hedonic scale (Appendix(ix))

Table 4.15. Result of sensory evaluation test on colour in pasteurized milk

Sample code	N	Median	Sum of rank
357	30	3.000	42.0
432	30	4.000	63.0
524	30	5.000	75.0

The result of effect on the smell of the pasteurized milk is showing the highest rank for the product ,which sample containing 40% chocolate liquid and 60% cocoa powder in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P= 0.132of the test is grater than minimum probability P=0.05. According to data sample chocolate liquid 40% and cocoa powder 60% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for smell. Therefore this sample (code no 432) come under the category of like very much according to the 7-point hedonic scale (Appendix(x))

Table 4.16. Result of sensory evaluation test on smell in pasteurized milk

Sample code	N	Median	Sum of rank
357	30	4.0000	55.0
432	30	5.0000	68.0
524	30	4.0000	56.5

The result of effect on the appearance of the pasteurized milk is showing the highest rank for the product ,which sample containing 40% chocolate liquid and 60% cocoa powder in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P= 0.109of the test is grater than minimum probability P 0.05. According to data sample chocolate liquid 40% and cocoa powder 60% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for appearance. Therefore this sample (code no 432)come under the category of like very much according to the 7-point hedonic scale (Appendix.(xi))

Table 4.17. Result of sensory evaluation test on appearance in pasteurized milk

Sample code	N	Median	Sum of rank
357	30	4.3333	51.5
432	30	5.0000	66.5
524	30	4.6667	62.0

The result of effect on the overall acceptability of the pasteurized milk is showing the highest rank for the product .which sample containing 60% chocolate liquid and 40% cocoa powder in the formula .According to data analysis there is a significant difference between the 3 samples, since probability value P 0.014of the test is less than minimum probability P 0.05. According to data sample chocolate liquid 60% and cocoa powder 40% with other ingredients remaining constant with gain the highest sum of rank value with the highest estimated median for overall acceptability. Therefore this sample (code no 524)come under the category of like very much according to the 7-point hedonic scale (Appendix(xii))

Table 4.18. Result of sensory evaluation test on overall acceptability in pasteurized milk

Sample code	N	Median	Sum of rank
357	30	4.2500	47.5
432	30	5.0833	64.5
524	30	5.9167	68.0

Finally the product specification of three types of in-container sterilized milk (Appendix (xiv)) and three types of pasteurized milk (Appendix (xv)) were estimate

Table 4.19. Results for modifying formulas pasteurized and sterilized milk

Product	Fat(%)	Acidity	Total Solids(%)
Sterilized milk			
436	2.0	0.15	17.6
542	2.0	0.15	17.6
678	2.0	0.15	17.6
Pasteurized milk			
357	2.1	0.15	17.8
432	2.1	0.15	17.8
524	2.1	0.15	17.8

The results of microbiological tests of pasteurized milk were evaluated.Total plate count results observed following as.

After the 48hrs incubation, number of the colonies appeared in Petridis standard method ager .All colonies were counted in Petridis and TPC was counted an according to the SLS standards, as follows.

Number of microbial cell per ml = number of colonies *dilution factor

Colonies per plate X

Dilution factor 10^{Y}

Volume of distribution added to the plate =1ml

So microbial content $X*10^{Y}$ cell /1ml

The amount of colonies appeared in the Petridish after 48hrs

Table 4.20 The amount of colonies

Microbial count	Added dilution to plate	No of colonies per pleat	Microbial count 1g of sample
10^{-1}	1ml	No detected	-
10^{-2}	1ml	No detected	-
10^{-3}	1ml	No detected	-
10^{-4}	1ml	3	$3*10^{4}$

The total plat count of the product can be identified as $3*10^{4}$ cells per 1ml which shows least number of viable cells among the slandered method Petridis.Methylene blue reduction was observed after 4 hours for each sample of pasteurized milk.

Chcolate liquid named "LiCo" was used to enhance other organoleptic qualities such as taste, smell, colour e.t.c.According to the statistically analyzed data shows code no 678 is best formula than other samples in overall acceptability.

Figure4.1. Initial product

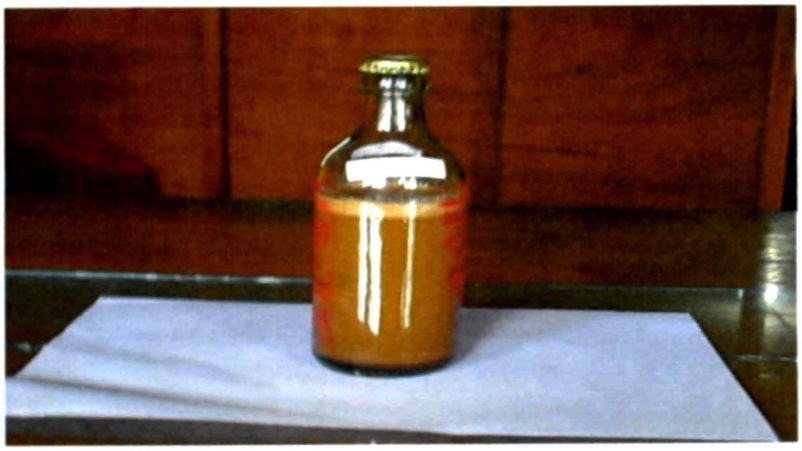


Figure4.2.Code no 436

Figure4.3.Code no 542

Figure4.4. Code no 678

Figure4.5. Initial product and modified 3 types of in container sterilized milk

The reduction in sediment achieved was approximately 65%in sample code no678.(Figure4.5)However other sample had less sediment percentage than code no678.Sediment percentage of them are 7%and 10.5%for code no 436(Figure4.2) and542(Figure4.3) respectivly. Sample code no678 (Figure4.4) had 16% of reduction sediment.According to the analyzed result most of untrained panelist like very much to sample code no 678 with considering other organoleptic qualities.So, the one containing chocolate liquid 30% and cocoa powder 70% is the most suitable ingredient combination than other formula for pasteurized milk.

According to the ballet paper mentioned result ,unknown panelists mentioned, that some like in-container sterilized milk for they like milk flavour than chocolate flavor as they containing chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01%. It cause colour appeared to be in light brown than others.

According to data sample chocolate liquid 30% and cocoa powder 70%,with a level of Carrageenan at 0.01% with other ingredients remaining constant which gained the highest sum of rank value with the highest estimated median for taste, colour,smell sedimentation and overall acceptability. Therefore this sample code no 678 comes under the category - "like very much" according to the 7-point hedonic scale

According to the sample data, 60% of chocolate liquid and 40% of cocoa powder with other ingredients remaining constant, gaining the highest sum of rank value with the highest estimated median for overall acceptability. Therefore this sample (code no 524) comes under the category - like very much according to the 7-point hedonic scale.

Total solids percents were not changed and it matched with the company product specification. Total solids percent of in container sterilized milk (Appendix (xiv)) was 17.6%.This value is slightly high than the product total solid-17.5%.The value can be fluctuated with in the range between 17.0% to 18.0. % according to the company specifications. Hence 17.6% of total solid is not a problem with the product specification.Pasteurized milk(Appendix (xv)) also contained 17.8% of total solids. It also lies within the range of product specifications; from17.0% to18.0%.Fat percentage in modified pastueized milk product was 2.1%. The maximum fat percentage should be contained in pasteurized milk is 2.1%.(Appendix (xv)) Hence this product is also acceptable. Acidity, Methylene blue reduction time and total colony count were 0.15, 4 hours and 30,000 per /ml respectively in modified pasturized milk product.

The product specification of earlier mentioned product was not changed by the usage of stabilizers. Hence the 0.01 percentage of stabilizer can be recommended for the application.

The shelf life of the pasteurized milk results revealed the product is acceptable for 4 days. Generally the pasteurized milk available in market level shelf life was also 4 days .Total plat count of product was 30000 per 1ml .It was below the maximum acceptable level in the .presence of microorganisms due to contamination during preparation. Normally market in container sterilized milk has 6 month shelf life. Hence the shelf life of modified products could not be observed within the short research duration.

CHAPTER 05

CONCLUSION AND RECOMMENDATION

5.1. Conclusions

According to the observations and results, final conclusions can be expressed as follow;

(a) According to the product specification Carrageenan 0.01% was selected as suitable amount to effectively reduce the sediment in in-container sterilized milk , without affecting the sensory attributes.

(b)According to the results the best sample of in-container sterilized milk was selected as the one containing chocolate liquid 30% and cocoa powder 70%, with a level of Carrageenan at 0.01% with other ingredients remaining constant.

(c) The sample containing 20% chocolate liquid and 80% cocoa powder was selected as the best one in pasteurized milk.

(d) The reduction in sediment achieved was approximately 65% in in-container sterilized milk

5.2. Recommendations

- Further reduction of sedimentation altering the product quality by using natural agents could be studied.
- Possibilities for replacing Carrageenan with a low- cost, natural stabilizer could also be studied further

REFERENCES

Fox, P.F. and McSweeney, P.L.H. (1998) Dairy Chemistry, and Microbiology, Blackie Academic and Professional, 1st edition, 478p.

FAO Regional Dairy Development and Training Centre for Asia and the Pacific, (1975)Milk Processing Dairy Training and Research Institute. University of Philippines ,Laguna.83p

Gorden, J. (1997) Dairy products, 74-138pp, In: Food Industries Manual, 24th edition, Edited by Ranken, M.D., Kill, R.C. and Baker, C.G.J. Blackie Academic and Professional, 650p.

Antherton V.H.,Newlander J.A.(1987),Chemistry and Testing of Dairy Products,4th edition,CBS publishers and distributers,309p.

Lewis, M.J. (1993) Physical Properties of Dairy Products, 331-380pp, In: Modern Dairy Technology, Volume 2, 2nd edition, Edited by Robinson, R.K., Blackie Academic and Professional, 516p.

Roland, P.C., David, H.L., and Terry, A.H. (2000) Guidelines for Sensory Analysis in Food Product Development and Quality Control, Aspen publishers Inc., 2nd edition, 210p.

Marnam, A.H., Sutherland, J.P. 2001. Milk and milk products: Technology, chemistry and microbiology. Aspen Publishers Inc., 451p.

Desilva.A.M.Y.J.(1991)The Technology on Milk and Milk Product, published by Industrial Technology Information Services of the Industrial Development Board.108p.

Alford ,Johnson.W.(1987) Fundermental of Dairy Chemistry,Published by S.K.Tain for CBC publishers, 928p.

CombsW.S. , Eckles(1976) ,Milk and Milk Products, Tata Mcgaw Hill publishing company Ltd.699p.

Burton, Cithienlin H. and Pien J.(1965),Milk Sterilization, Food and Agreculture Organisation of The United nation,456p.

Robison K.R.(1993) Modern Dairy Technology, John Wiley and Sons Inc.publication, 516p.

Sri Lanka Standard Institution (1988) Methods of test for milk and milk products, SLS 735-Part 4:Determination of Acidity, 7p.

Sri Lanka Standard Institution (1987) Methods of milk and milk products,SLS 735-Part 3:Determination of moisture, 6p.

Sri Lanka Standard Institution (1991)Specification of milk added drinks, SLS 917Requirments, 8p.

Sri Lanka Standard Institution (1986)Chocolate, SLS 326:Definitions, 21p.

Sri Lanka Standard Institution (1987) Methods of test for milk and milk products,SLS 735-Part 2:Determination of titrtable acidity, 17p.

MCPI Corperation . IntroductionNatural Grade Carrageenan (available from: http://carrageenan.cebu.ph/carrageenan.html accessed on:10/11/2008)

Martine Chaplin(2008), LondonSouth Bank University,Carrageenan(available from: http://www.lsbu.ac.uk/water/hycar.html accessed on:06/12/2008)

APPENDIX I

Sabaragamuwa University of Sri Lanka
Faculty of Applied Sciences, BSc. Food Science & Technology
Department of Food Science & Technology

Questionnaire for Sensory Analysis (Seven Point Hedonoic Test)

Product: **Sterilized milk**

Date: ..... Time : .....

- Assess the sample individually.
- Indicate how much you preferred each sample after testing.
- Rinse you mouth with water after tasting each sample.
- Give numerical values ranking from Like very much to Dislike very much.

Point Scale	Points
Like very much	7
Like moderately	6
Like slightly	5
Neither like nor dislike	4
Dislike slightly	3
Dislike moderately	2
Dislike very much	1

Sensory Aspects	Sample code		
	436	542	678
Taste			
Colour			
Smell			
Appearance(sedimentation)			
Overall Acceptability			

Comments:

.....

.....

.....

.....

.....

.....

Thank you

Signature

APPENDIX II

Sabaragamuwa University of Sri Lanka
Faculty of Applied Sciences, BSc. Food Science & Technology
Department of Food Science & Technology

Questionnaire for Sensory Analysis (Seven Point Hedonoic Test)

Product: Pasteurized milk

Date: ..... Time : .....

- Assess the sample individually.
- Indicate how much you preferred each sample after testing.
- Rinse you mouth with water after tasting each sample.
- Give numerical values ranking from Like very much to Dislike very much.

Point Scale	Points
Like very much	7
Like moderately	6
Like slightly	5
Neither like nor dislike	4
Dislike slightly	3
Dislike moderately	2
Dislike very much	1

Sensory Aspects	Sample code		
	357	432	524
Taste			
Colour			
Smell			
Appearance			
Overall Acceptability			

Comments:

.....

.....

.....

.....

Thank you

Signature

APPENDIX III

Friedman Test: taste versus s blocked by code no

```
S = [illegible]   DF = [illegible]   P = 0.028
S = [illegible]   DF = [illegible]   P = 0.023 (adjusted for ties)

                                  Sum
                                   of
s        N   Est Median         Ranks
4[illegible]    [illegible]   [illegible]   49.0
54[illegible]   [illegible]   4.[illegible] [illegible]
[illegible]     [illegible]   4.[illegible] [illegible]

Grand median = 4.0000
```

Ho : There are not different between three in taste.

H_1 : There are different between three in taste.

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.023.

0.023<0.05. Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in taste

APPENDIX IV

Friedman Test: colour versus s blocked by code no

```
S = [illegible]   DF = [illegible]   P = 0.006
S = 11.[illegible]   DF = [illegible]   P = 0.00[illegible] (adjusted for ties)

                                    Sum
                                     of
s          N   Est Median         Ranks
[illegible]   [illegible]   [illegible]   [illegible]
[illegible]   [illegible]   [illegible]   [illegible]
[illegible]   [illegible]   4.[illegible]   [illegible]4.[illegible]

Grand median   [illegible]
```

Ho : There are not different between three in colour

H_1 : There are different between three in colour.

If p value less than alpha. Then Ho is rejected.

Statistically resulted p value is 0.003.

0.003< 0.05. Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in colour.

APPENDIX V

Friedman Test: smell versus s blocked by code no

```
S    6.67  DF   2  P   0.036
S    7.21  DF   2  P   0.027  (adjusted for ties)

                                 Sum
                                  of
          N  Est  Median   Ranks
436      40      4.1667    80.0
542      40      4.8333    60.0
678      40      5.5000   100.0

Grand median    4.8333
```

Ho : There are not different between three in smell

H_1 : There are different between three in smell.

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.027

0.027<0.05 Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in smell.

APPENDIX VI

Friedman Test: sedimentation versus s blocked by code no

```
S    8.72  DF    2  P    0.013
S    9.60  DF    2  P    0.008 (adjusted for ties)

                      Sum
                Est    of
S       N  Median  Ranks
4.16   40   4.000   81.5
5.47   40   5.000   95.5
6.78   40   6.000   73.0

Grand median   5.000
```

Ho : There are not different between three in sedimentation

H_1 : There are different between three in sedimentation

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.008.

0.008<0.05 Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in sedimentation

APPENDIX VII

Friedman Test: overall acceptability versus s blocked by code no

```
S = 24.72  DF = 2  P = 0.000
S = 27.08  DF = 2  P = 0.000 (adjusted for ties)

                        Sum
                Est      of
s       N  Median   Ranks
436    40   5.000    41.0
542    40   6.000    59.5
678    40   7.000    79.5

Grand median    6.000
```

Ho : There are not different between three in overall acceptability

H_1 : There are different between three in overall acceptability

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.000

0.000<0.05 Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in overall acceptability

APPENDIX VIII

Friedman Test: Taste versus s blocked by code no

S = 15.82 DF = 2 P = 0.000
S = 16.65 DF = [illegible] P = 0.000 (adjusted for ties)

s	N	Est Median	Sum of Ranks
357	30	3.000	43.5
432	30	4.000	[illegible]
524	30	5.000	74.0

Grand median = 4.000

Ho : There are not different between three in taste

H_1 : There are different between three in taste

If p value less than alpha. Then Ho is rejected.

Statistically resulted p value is 0.000

0.000<0.05 Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in taste

APPENDIX IX

Friedman Test: Colour versus s blocked by code no

```
      [illegible]  DF    2  P    0.000
      [illegible]  DF    1  P    0.000 (adjusted for ties)

                              Sum
                       Est     of
          N  Median   Median  Ranks
[illegible]  40  [illegible]  4.000  [illegible]
[illegible]  40  [illegible]  4.000  [illegible]
[illegible]  40  [illegible]  5.000  [illegible]

Grand median    4.000
```

Ho : There are not different between three colour

H_1 : There are different between three in colour

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.000

0.000< 0.05 Then Ho is rejected.

Finally can be concluded as that there are difference between three samples in colour

APPENDIX X

Friedman Test: Smell versus s blocked by code no

```
S    1.65  DF   2  P  0.161
S    4.06  DF   2  P  0.132 (adjusted for ties)

                              Sum
                               of
S       N  Est  Median      Ranks
3.57   10       4.0000      55.0
4.43   10       5.0000      68.5
5.24   10       4.0000      56.5

Grand median    4.3333
```

Ho : There are not different between three in smell

H_1 : There are different between three in smell

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.132

0.132>0.05 Then Ho is not rejected.

Finally can be concluded as that there are not difference between three samples in smell

APPENDIX XI

Friedman Test: Appearance versus s blocked by code no

```
S     4.95  DF   2  P   0.139
S     4.43  DF   2  P   0.109 (adjusted for ties)

                              Sum
                               of
s        N  Est  Median    Ranks
3.7     30       4.3333    51.5
4.2     30       5.0000    66.5
5.4     30       4.6667    62.0

Grand median    4.6667
```

Ho : There are not different between three in appearance.

H_1 : There are different between three in appearance

If p value less than alpha, Then Ho is rejected.

Statistically resulted p value is 0.109

0.109>0.05 Then Ho is not rejected.

Finally can be concluded as that there are not difference between three samples in appearance.

APPENDIX XII

Friedman Test: Overall Acceptability versus s blocked by code no

```
S = 8.02   DF = 2   P = 0.018
S = 8.51   DF = 2   P = 0.014 (adjusted for ties)

                        Sum
                         of
s      N   Est Median   Ranks
357   30        4.2500   47.5
432   30        5.0833   64.5
524   30        5.9167   68.0

Grand median = 5.0833
```

Ho : There are not different between three in overall acceptability.

H_1 : There are different between three in overall acceptability

If p value less than alpha, Then Ho is rejected.

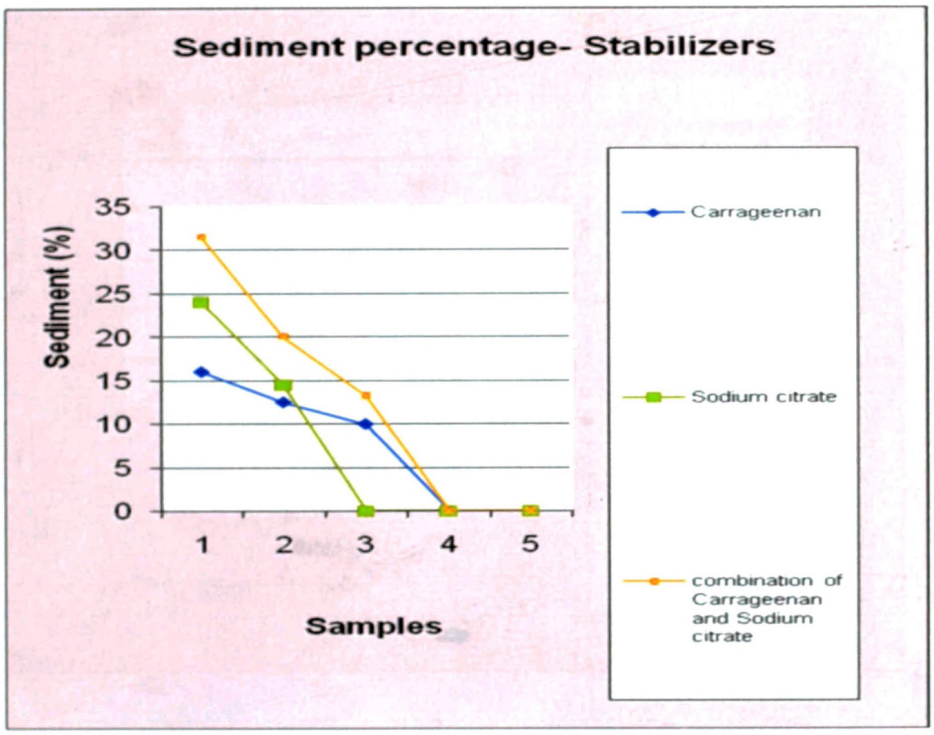
Statistically resulted p value is 0.014

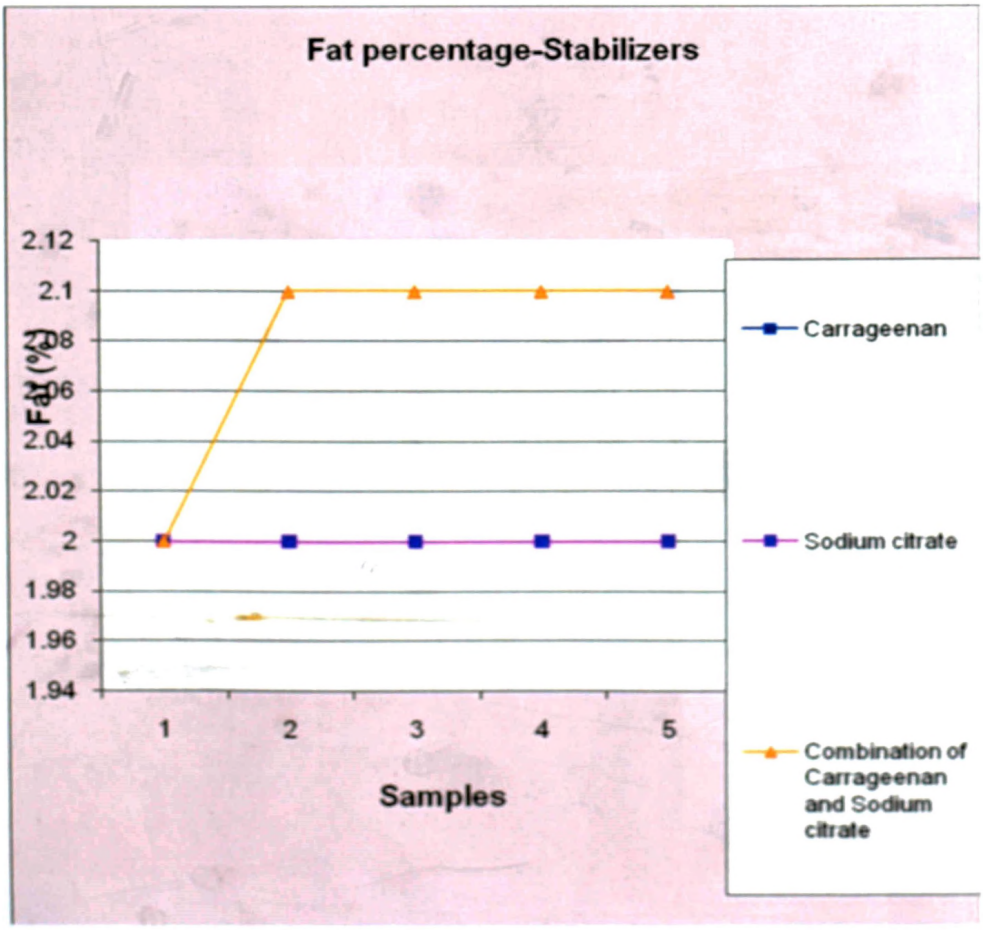
0.014<0.05 Then Ho is rejected.

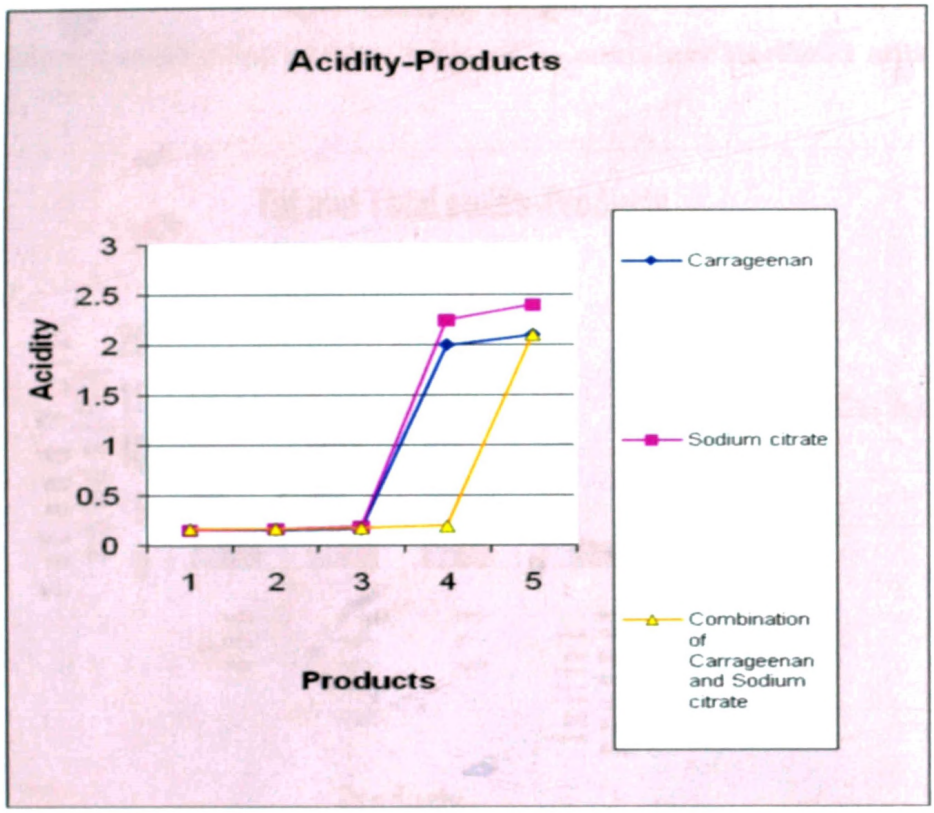
Finally can be concluded as that there are difference between three samples in overall acceptability.

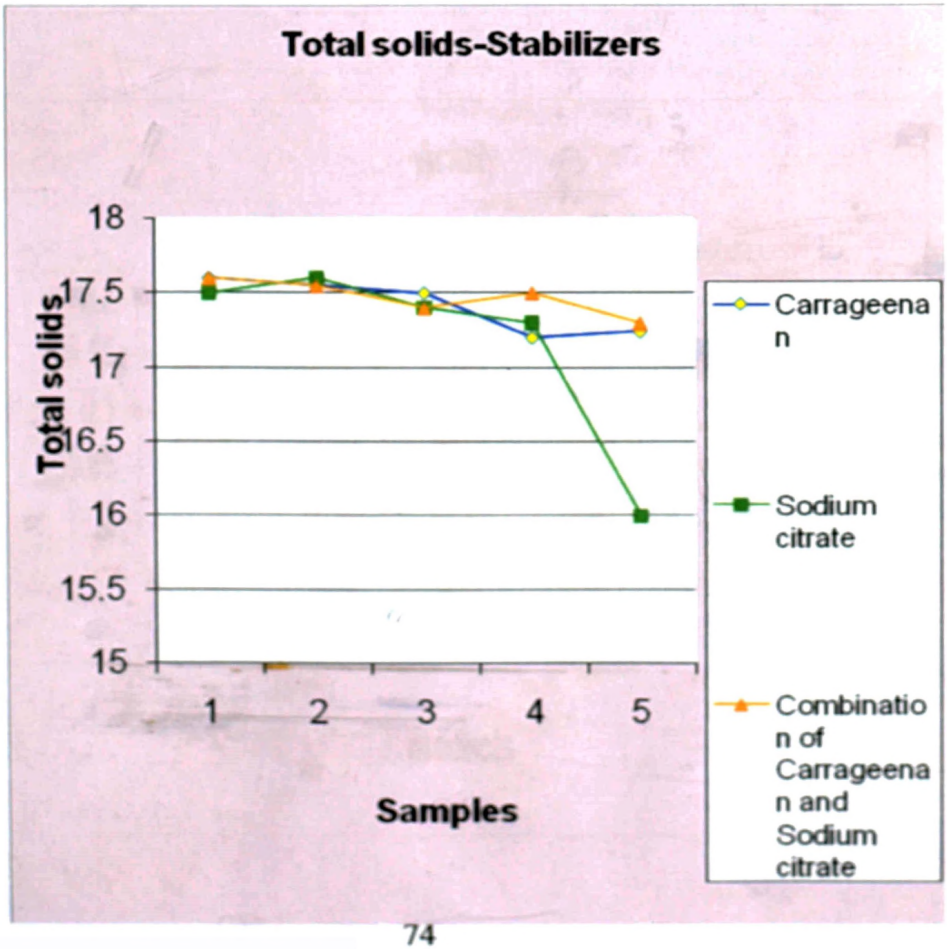
APPENDIX XIII

Selection of best stabilizer appropriate amount -in container sterilized milk



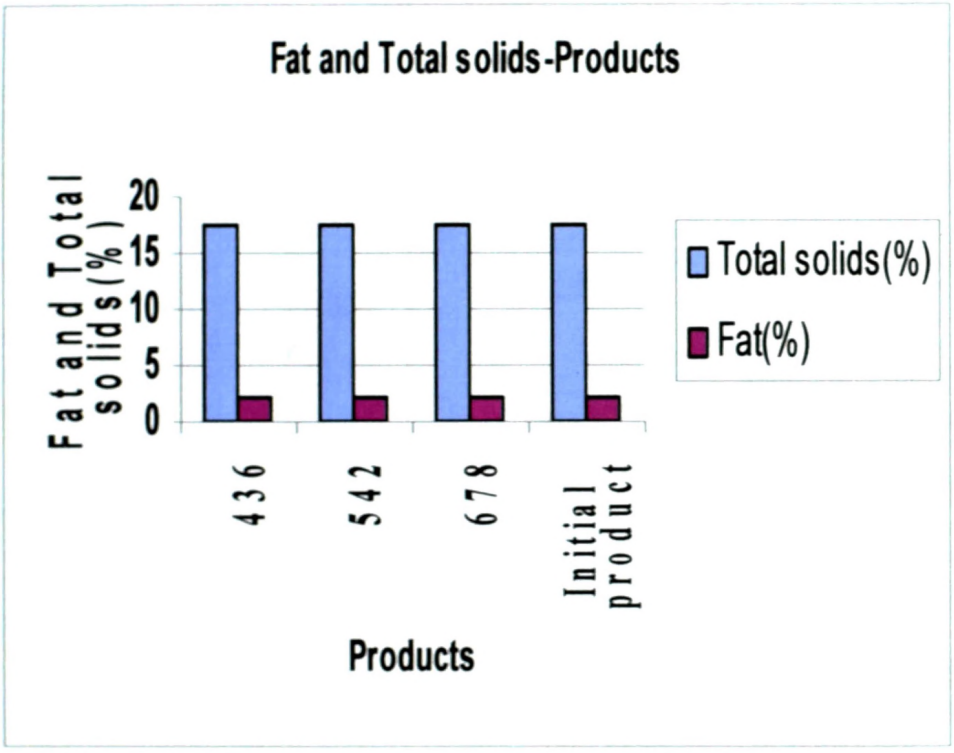


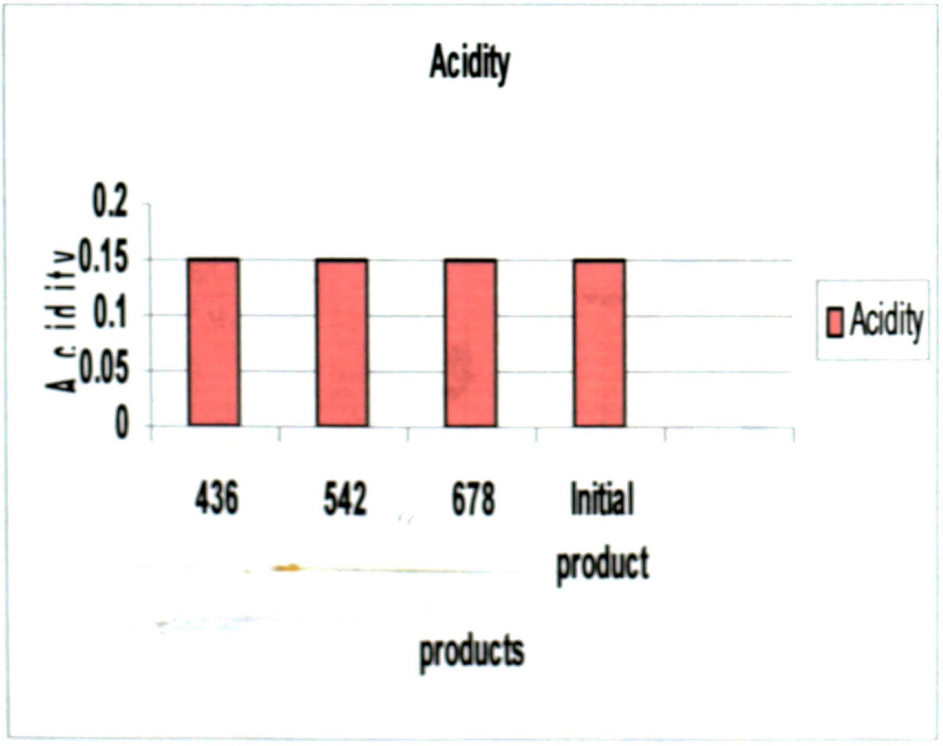

Acidity-Products
Acidity
3
2.5
2
1.5
1
0.5
0
1
2
3
4
5
Products
Carrageenan
Sodium citrate
Combination of Carrageenan and Sodium citrate


Total solids-Stabilizers
Total solids
18
17.5
17
16.5
16
15.5
15
1
2
3
4
5
Samples
Carrageenan
Sodium citrate
Combination of Carrageenan and Sodium citrate

APPENDIX XIV

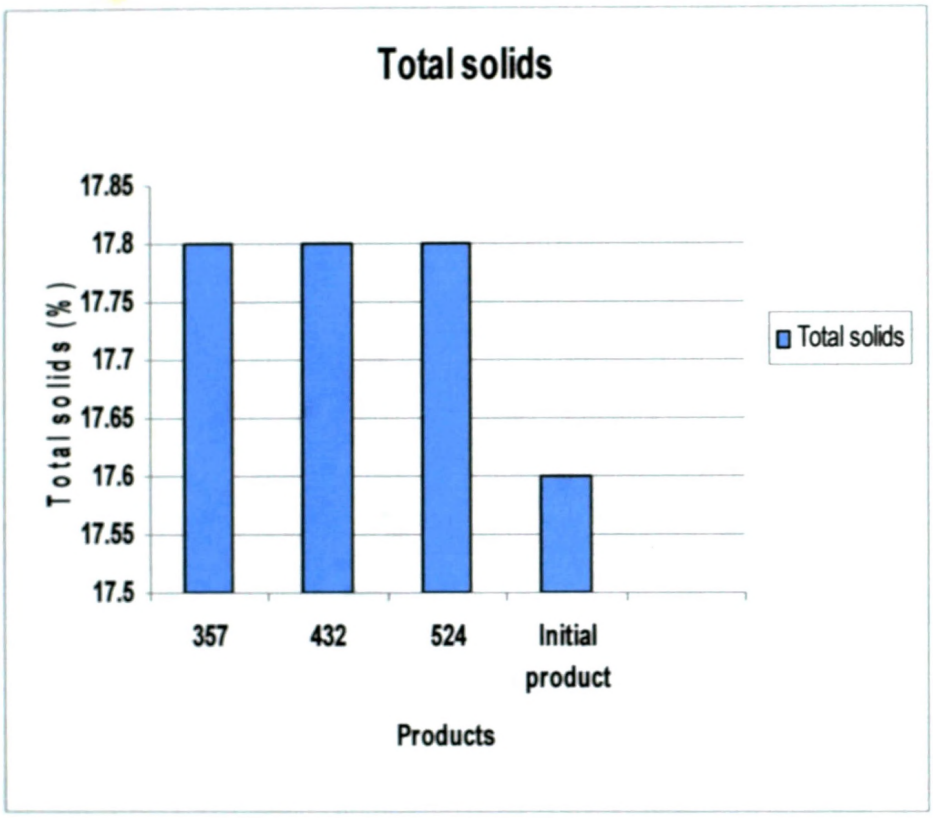
The product specification of three types of in- container sterilized milk

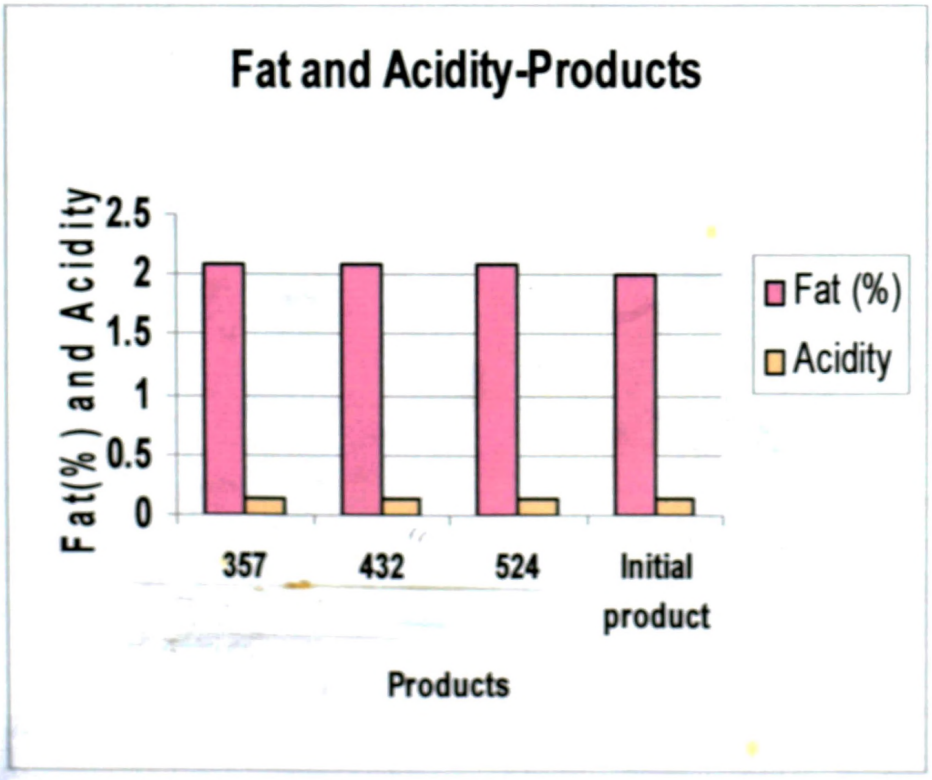




APPENDIX XV

The product specification of three types of pasteurized milk





National Digitization Project

National Science Foundation

Institute : Sabaragamuwa University of Sri Lanka

1. Place of Scanning : Sabaragamuwa University of Sri Lanka, Belihuloya
2. Date Scanned : ..2017-09-19.....
3. Name of Digitizing Company : Sanje (Private) Ltd, No 435/16, Kottawa Rd, Hokandara North, Arangala, Hokandara
4. Scanning Officer

Name : ...S.A.C. Sadaruwan.....

Signature : .....

Certification of Scanning

I hereby certify that the scanning of this document was carried out under my supervision, according to the norms and standards of digital scanning accurately, also keeping with the originality of the original document to be accepted in a court of law.

Certifying Officer

Designation : .....Librarian.....

Name : .....T. N. Neighsoorei.....

Signature : .....

MR. T.N. NEIGHSOOREI
Librarian
Sab. ...

Date : ..2017-09-19.....